Supplementary information

Synthesis and evaluation of anticancer activity of new 9-acridinyl amino acid derivatives

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Physicochemical characterization data of synthesized compounds

(S)-methyl 2-(acridin-9-ylamino)-3-phenylpropanoate (1). Compound **1** was synthesized according to the general procedure, using sodium methoxide solution and L-phenyalanine methyl ester hydrochloride. This derivative was purified using preparative thin layer chromatography with mobile phases ethyl acetate/hexane 4:0.3 (v/v) and chloroform/methanol 9:1.6 (v/v). Yellow crystalline solid was obtained after recrystallization in diethyl ether. Yield 21%. Mp 176.0-179.5 °C. IR (ATR) ν = 705.0, 789.20, 1209.67, 1470.37, 1587.16, 1739.65, 3236.92 cm^{-1}. ^{1}H NMR (400 MHz, $CDCl_3$) δ 8.26 (d, 2H, J = 6.8 Hz, H-4 and H-5), 7.96 (d, 2H, J = 8.8 Hz, H-1 and H-8), 7.71 (t, 2H, J = 7.6 Hz, H-3 and H-6), 7.39 (t, 2H, J = 8 Hz, H-2 and H-7), 7.20-7.31 (m, 5H, H-4', H-5', H-6', H-7' and H-8'), 5.03 (s, 1H, H-1'), 3.64 (s, 3H, $-CH_3$), 3.40 (d, 2H, H-2'). ^{13}C NMR (100 MHz, $CDCl_3$) δ 135.31, 131.44, 129.55, 128.80, 127.64, 124.47, 122.59, 77.34, 77.22, 77.02, 76.70, 70.62, 62.73, 52.63, 40.13, 29.71. m/z = 357.4 (M^{+}+1), 206.18, 179.19, 297.26. MS [M+H]+ calculated for C23H20N2O2 = 357.15248; observed = 357.15297.

(S)-methyl 2-(acridin-9-ylamino)-3-(1H-imidazol-4-yl)propanoate (2). Compound **2** was synthesized according to the general procedure, using sodium methoxide solution and L-histidine methyl ester dihydrochloride. The reaction mixture was purified using preparative thin layer chromatography with mobile phase chloroform/methanol 9:1.6 (v/v). Yellow crystalline solid was obtained after recrystallization in diethyl ether. Yield: 77%. Mp 95-97.5 °C. IR (ATR) ν = 750.67, 1168.29, 1205.05, 1473.66, 1636.99, 1738.11, 2872.03 cm^{-1}. ^{1}H NMR (400 MHz, $CDCl_3$) δ 8.25 (d, 4H, J = 8.4 Hz, H-1, H-4, H-5 and H-8), 7.63-7.69 (m, 3H, H-3, H-6 and H-5'), 7.37 (t, 2H, J=7.6 Hz, H-2 and H-7), 6.95 (s, 1H, H-7'), 5.28 (t, 1H, J=4.4 Hz, H-1'), 3.68 (s, 3H, $-CH_3$), 3.30-3.49 (m, 2H, H-2'). ^{13}C NMR (100 MHz, $CDCl_3$) δ 171.30, 154.80, 143.30, 135.65, 133.00, 124.11, 123.90, 123.36, 114.76, 61.81, 52.98, 30.76.

m/z = 347.1 (M^++1), 205.98, 178.99, 234.96. MS [M+H]+ calculated for C20H19N4O2 = 347.14298; observed = 347.14966.

(S)-methyl 2-(acridin-9-ylamino)-3-(1-methyl-1H-indol-3-yl)propanoate (3). Compound **3** was synthesized according to the general procedure, using sodium methoxide solution and 1-methyl-L-tryptophan methyl ester hydrochloride. The reaction mixture was purified using preparative thin layer chromatography with mobile phases chloroform/methanol 9:1.6 (v/v) and ethyl acetate/hexane 4:1 (v/v). Yellow crystalline solid was obtained after recrystallization in diethyl ether. Yield: 31%. Mp 55-58.5 °C. IR (ATR) ν = 738.83, 1205.01, 1441.65, 1470.75, 1525.13, 1557.74, 1737.67 cm^{-1}. ^1H NMR (400 MHz, DMSO) δ ppm 8.12-8.15 (m, 2H, H-1 and H-8), 7.51-7.74 (m, 6H, H-2, H-3, H-4, H-5, H-6 and H-7), 7.33 (d, 1H, J=8 Hz, H-10'), 7.09-7.15 (m, 3H, H-4', H-7', H-8'), 6.95 (t, 1H, J=7.6 Hz, H-9'), 5.02 (s, 1H, H-1'), 3.64 (s, 3H, -CH_3), 3.54 (s, 3H, H-12'), 3.49-3.50 (m, 2H, H-2'). ^{13}C NMR (100MHz, DMSO), δ 173.12, 136.95, 131.16, 129.00, 127.87, 121.54, 119.01, 118.85, 110.03, 52.41, 32.64. m/z = 410.2 (M^++1), 174.06, 216.04, 144.07. MS [M+H]+ calculated for C26H24O2N3 = 410.17903; observed = 410.18594.

(S)-2-(acridin-9-ylamino)-3-(1-methyl-1H-indol-3-yl)propanoic acid (4). Compound **4** was synthesized according to the general procedure, using sodium methoxide solution and 1-methyl-L-tryptophan. The reaction mixture was purified using preparative thin layer chromatography with mobile phase chloroform/methanol 9:2 (v/v). Orange crystalline solid was obtained after recrystallization in chloroform/methanol 5:5 (v/v). Yield: 57%. Mp 43-46.5 °C. IR (ATR) ν = 737.28, 1271.24, 1370.64, 1529.12, 1586.03, 1634.44, 2850.80, 2920.97. cm^{-1}. ^1H NMR (400 MHz, CD_3OD) δ 8.01 (d, 2H, J = 8.4 Hz, H-1 and H-8), 7.63 (t, 2H, J=8 Hz, H-3 and H-6), 7.40 (d, 2H, J=8.4 Hz, H-4 and H-5), 7.17 (t, 2H, J=7.6 Hz, H-2 and H-7), 6.98 (d, 1H, J=7.6 Hz, H-10'), 6.83-6.92 (m, 2H, H-7' and H-8'), 6.68 (s, 1H, H-4'), 6.52 (t, 1H, J=7.2 Hz, H-9'), 5.23 (s, 1H, H-1'), 3.55-3.59 (m, 2H, H-2'), 3.30 (s, 3H, H-

12'). ^{13}C NMR (100 MHz, CD_3OD) δ 156.98, 139.09, 136.58, 134.39, 130.95, 128.17, 127.27, 123.16, 120.96, 118.43, 117.94, 117.69, 112.39, 108.59, 108.53, 64.28, 30.99, 29.34. m/z = 396.1 (M^++1), 195.05, 144.06, 202.03. MS [M+H]+ calculated for C25H22O2N3 = 396.16338; observed = 396.17023.

Methyl 8-(acridin-9-ylamino)octanoate (5). Compound **5** was synthesized according to the general procedure, using sodium methoxide solution and 8-aminooctanoic acid methyl ester hydrochloride. The reaction mixture was purified using preparative thin layer chromatography with mobile phase chloroform/methanol 9:1.6 (v/v). Yellow crystalline solid was obtained after recrystallization in diethyl ether. Yield: 38%. Mp 106.3-109.0 °C. IR (ATR) ν = 662.28, 748.69, 1165.98, 1466.81, 1566.82, 1633.69, 1734.35, 2738.24 cm^{-1}. ^{1}H NMR (400 MHz, $CDCl_3$) δ 8.18 (d, 2H, J=8 Hz, H-4 and H-5), 7.95 (d, 2H, J=8 Hz, H-1 and H-8), 7.41 (d, 2H, J=7.2 Hz, H-3 and H-6), 7.17 (t, 2H, J=8 Hz, H-2 and H-7), 4.04 (t, 2H, J=7.6 Hz, 2×H-1'), 3.67 (s, 3H, 3×H-8'), 2.39 (t, 2H, J=7.2 Hz, 2×H-7'), 2.02 (quin, 2H, J=7.2 Hz, 2×H-6'), 1.55-1.66 (m, 4H, 2×H-2' and 2×H-3'), 1.38-1.46 (m, 4H, 2×H-4' and 2×H-5'). ^{13}C NMR (100 MHz, $CDCl_3$) δ 174.18, 155.70, 141.16, 132.92, 124.75, 122.80, 121.04, 112.91, 51.48, 48.75, 33.97, 30.55, 28.97, 28.94, 26.76, 24.77. m/z = 351.2 (M^++1), 195.03, 150.99, 178.01. MS [M+H]+ calculated for C22H27O2N2 = 351.19943; observed = 351.20621.

Ethyl 4-(acridin-9-ylamino)butanoate (6). Compound **6** was synthesized according to the general procedure, using sodium ethoxide solution and 4-aminobutyric acid ethyl ester hydrochloride. The reaction mixture was purified using preparative thin layer chromatography with mobile phase chloroform/methanol 9:1.6 (v/v). Yellow crystalline solid was obtained after recrystallization in diethyl ether. Yield: 36%. Mp 148-150.5 °C. IR (ATR) ν_{max} (cm^{-1}): 747.07, 1171.20, 1477.01, 1569.49, 1588.35, 1635.85, 1725.72, 2779.74. ^{1}H NMR (400 MHz, DMSO) δ ppm 8.58 (d, 2H, J = 8 Hz, H-4 and H-5), 7.92 (s, 4H, H-1, H-3, H-6 and H-8), 7.52 (d, 2H, J = 6.4, H-2 and H-7), 4.10 (2H, s, 2×H-4'), 4.00 (2H, q, J = 7.2 Hz, 2×H-1'), 2.13-

2.17 (m, 2H, 2×H-2'), 1.12 (3H, t, J=6.8, 3×H-5'). ^{13}C NMR (100 MHz, DMSO), δ ppm 172.98, 157.40, 140.78, 134.90, 126.36, 123.57, 119.57, 113.25, 60.43, 31.45, 31.18, 24.90, 14.49. m/z = 309.1 (M^++1), 87.10, 195.02, 281.01. MS [M+H]+ calculated for C19H21O2N2 = 309.15248; observed = 309.15955.

Propyl 4-(acridin-9-ylamino)butanoate (7). Compound **7** was synthesized according to the general procedure, using sodium propoxide solution and 4-aminobutyric acid propyl ester hydrochloride. The reaction mixture was purified using preparative thin layer chromatography with mobile phases ethyl acetate/hexane 4:1 (v/v) and chloroform/methanol 9:1 (v/v). Yellow crystalline solid was obtained after recrystallization in diethyl ether. Yield: 42%. Mp 156.5-159.8 °C. IR (ATR) ν = 664.40, 745.42, 939.82, 1172.32, 1467.45, 1588.44, 1635.45, 1730.53, 2778.87 cm^{-1}. ^{1}H NMR (400 MHz, $CDCl_3$) δ 8.28 (d, 2H, J=8.8 Hz, H-4 and H-5), 8.09 (d, 2H, J=8.4 Hz, H-1 and H-8), 7.49 (t, 2H, J=7.2 Hz, H-3 and H-6), 7.21-7.25 (m, 2H, H-2 and H-7), 4.22 (t, 2H, J=6.8 Hz, 2×H-4'), 4.11 (t, 2H, J=6.8 Hz, 2×H-1'), 2.73 (t, 2H, J=6.4 Hz, 2×H-3'), 2.36 (quin, 2H, J=6.4 Hz, 2×H-2'), 1.63-1.70 (m, 2H, 2×H-5'), 0.95 (t, 3H, J=7.2 Hz, 3×H-6'). ^{13}C NMR (100 MHz, $CDCl_3$) δ 174.15, 133.70, 124.89, 123.13, 119.98, 112.30, 66.83, 48.72, 32.00, 24.87, 21.94, 10.37. m/z = 323.2 (M^++1), 87.11, 195.07, 281.11. MS [M+H]+ calculated for C20H23O2N2 = 323.16813; observed = 323.17508.

Ethyl 3-(acridin-9-ylamino)propanoate (8). Compound **8** was synthesized according to the general procedure, using sodium ethoxide solution and β-alanine ethyl ester hydrochloride. The reaction mixture was purified using preparative thin layer chromatography with mobile phase chloroform/ethanol 9:1.6 (v/v). Yellow crystalline solid was obtained after recrystallization in diethyl ether. Yield: 29%. Mp 29.7-32.5 °C. IR (ATR) ν = 741.88, 1019.40, 1186.82, 1514.04, 1725.16, 3317.52 cm^{-1}. ^{1}H NMR (400 MHz, $CDCl_3$) δ 8.16 (d, 2H, J=8.4 Hz, H-4 and H-5), 8.08 (d, 2H, J=8.4 Hz, H-1 and H-8), 7.63 (t, 2H, J=7.2 Hz, H-3 and H-6), 7.36 (t, 2H, J=7.2 Hz, H-2 and H-7), 4.21 (q, 2H, J=7.2 Hz, H-3'), 4.11 (t, 2H, J=6

Hz, H-1'), 2.81 (t, 2H, J=6 Hz, H-2'), 1.27 (t, 3H, J=7.2 Hz, H-4'). ^{13}C NMR (100 MHz, $CDCl_3$) δ 172.44, 152.50, 146.88, 130.97, 127.07, 123.61, 123.34, 116.76, 61.14, 45.89, 34.90, 14.17. m/z = 295.2 (M^++1), 207.07, 206.06, 267.08. MS [M+H]+ calculated for C18H19O2N2 = 295.13683; observed = 295.14413.

Propyl 3-(acridin-9-ylamino)propanoate (9). Compound **9** was synthesized according to the general procedure, using sodium propoxide solution and β-alanine propyl ester hydrochloride. The reaction mixture was purified using preparative thin layer chromatography with mobile phase chloroform/propanol 9:1.6 (v/v). Yellow crystalline solid was obtained after recrystallization in diethyl ether. Yield: 22%. Mp 33-35.5 °C. IR (ATR) ν = 742.56, 753.98, 1139.08, 1183.86, 1512.75, 1562.24, 1723.97, 3311.60 cm^{-1}. ^{1}H NMR (400 MHz, $CDCl_3$) δ 8.14 (d, 2H, J=8.8 Hz, H-4 and H-5), 8.09 (d, 2H, J=8.4 Hz, H-1 and H-8), 7.66 (t, 2H, J=7.2 Hz, H-3 and H-6), 7.38 (t, 2H, J=7.2 Hz, H-2 and H-7), 4.06-4.14 (m, 4H, 2×H-1' and 2×H-3'), 2.77 (t, 2H, J=6 Hz, 2×H-2'), 1.64-1.69 (m, 2H, 2×H-4'), 0.94 (t, 3H, J=7.2 Hz, 3×H-5'). ^{13}C NMR (100 MHz, $CDCl_3$) δ 172.71, 151.97, 130.63, 127.97, 123.67, 123.09, 117.23, 66.75, 46.02, 34.90, 21.92, 10.35. m/z = 309.2 (M^++1), 207.06, 267.08, 179.07. MS [M+H]+ calculated for C19H21O2N2 = 309.15248; observed = 309.15948.

Ethyl 6-(acridin-9-ylamino)hexanoate (10). Compound **10** was synthesized according to the general procedure, using sodium ethoxide solution and 6-aminohexanoic acid ethyl ester hydrochloride. The reaction mixture was purified using preparative thin layer chromatography with mobile phase chloroform/methanol 9:1.6 (v/v). Yellow crystalline solid was obtained after recrystallization in diethyl ether. Yield: 33%. Mp 169.5-172.0 °C. IR (ATR) ν = 663.28, 751.97, 836.63, 868.97, 1173.36, 1468.92, 1529.01, 1566.20, 1587.08, 1634.41, 1723.38, 2804.29 cm^{-1}. ^{1}H NMR (400 MHz, DMSO-d6) δ 8.59 (d, 2H, J=6.8 Hz, H-4 and H-5), 7.93-7.99 (m, 4H, H-1, H-3, H-6 and H-8), 7.53 (t, 2H, J=6.8 Hz, H-2 and H-7), 3.99-4.09 (m, 4H, 2×H-1' and 2×H-6'), 2.29 (t, 2H, J=7.2 Hz, 2×H-5'), 1.89-1.92 (m, 2H, 2×H-4'), 1.56-1.59

(m, 2H, 2×H-2'), 1.30-1.45 (m, 2H, 2×H-3'), 1.14 (t, 3H, J=7.2 Hz, 3×H-7'). ^{13}C NMR (100 MHz, DMSO-d6) δ 173.21, 157.76, 135.26, 126.49, 123.71, 119.18, 60.13, 49.13, 33.77, 29.01, 26.10, 24.49, 14.57. m/z = 337.2 (M^++1), 195.06, 309.16, 69.26. MS [M+H]+ calculated for C21H25O2N2 = 337.18378; observed = 337.19083.

Propyl 6-(acridin-9-ylamino)hexanoate (11). Compound **11** was synthesized according to the general procedure, using sodium propoxide solution and 6-aminohexanoic acid propyl ester hydrochloride. The reaction mixture was purified using preparative thin layer chromatography with mobile phases ethyl acetate/hexane 4:1 (v/v) and chloroform/methanol 9:1.6 (v/v). Yellow crystalline solid was obtained after recrystallization in diethyl ether. Yield: 32%. Mp 145.7-149.3 °C. IR (ATR) ν = 662.50, 765.32, 1177.03, 1272.97, 1333.36, 1469.40, 1587.02, 1633.56, 1720.98, 2850.50 cm^{-1}. ^{1}H NMR (400 MHz, DMSO-d6) δ 8.55 (d, 2H, J=8.8 Hz, H-4 and H-5), 7.91-7.92 (m, 4H, H-1, H-3, H-6 and H-8), 7.50 (t, 2H, J=6.4 Hz, H-2 and H-7), 4.04 (t, 2H, J= 6.8 Hz, 2×H-6'), 3.93 (t, 2H, J=6.4 Hz, 2×H-1'), 2.30 (t, 2H, J=7.2 Hz, 2×H-5'), 1.87-1.90 (m, 2H, 2×H-4'), 1.51-1.59 (m, 4H, 2×H-2' and 2×H-7'), 1.37-1.41 (m, 2H, 2×H-3'), 0.84 (t, 3H, J=7.2 Hz, 3×H-8'). ^{13}C NMR (100 MHz, DMSO-d6) δ 173.28, 134.60, 126.35, 123.46, 120.11, 65.61, 49.36, 33.80, 29.30, 26.16, 24.56, 21.98, 10.69. m/z = 351.2 (M^++1), 195.00, 309.03, 69.26. MS [M+H]+ calculated for C22H27O2N2 = 351.19943; observed = 351.20612.

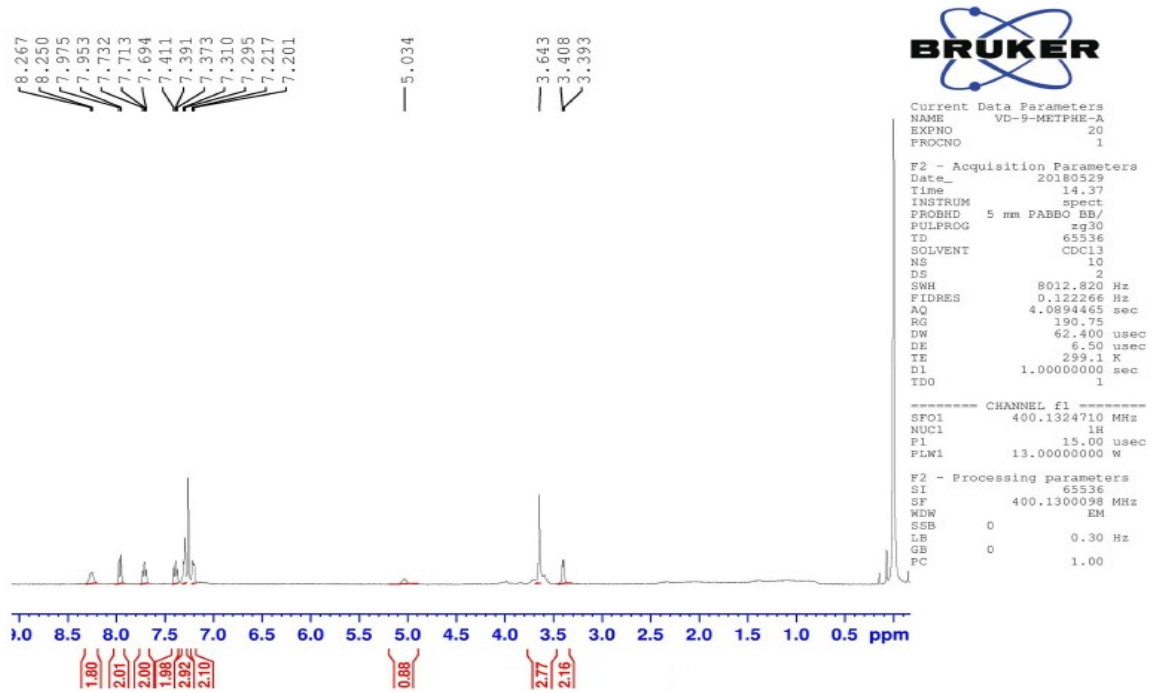


Figure S1. Compound 1 – ^{1}H NMR

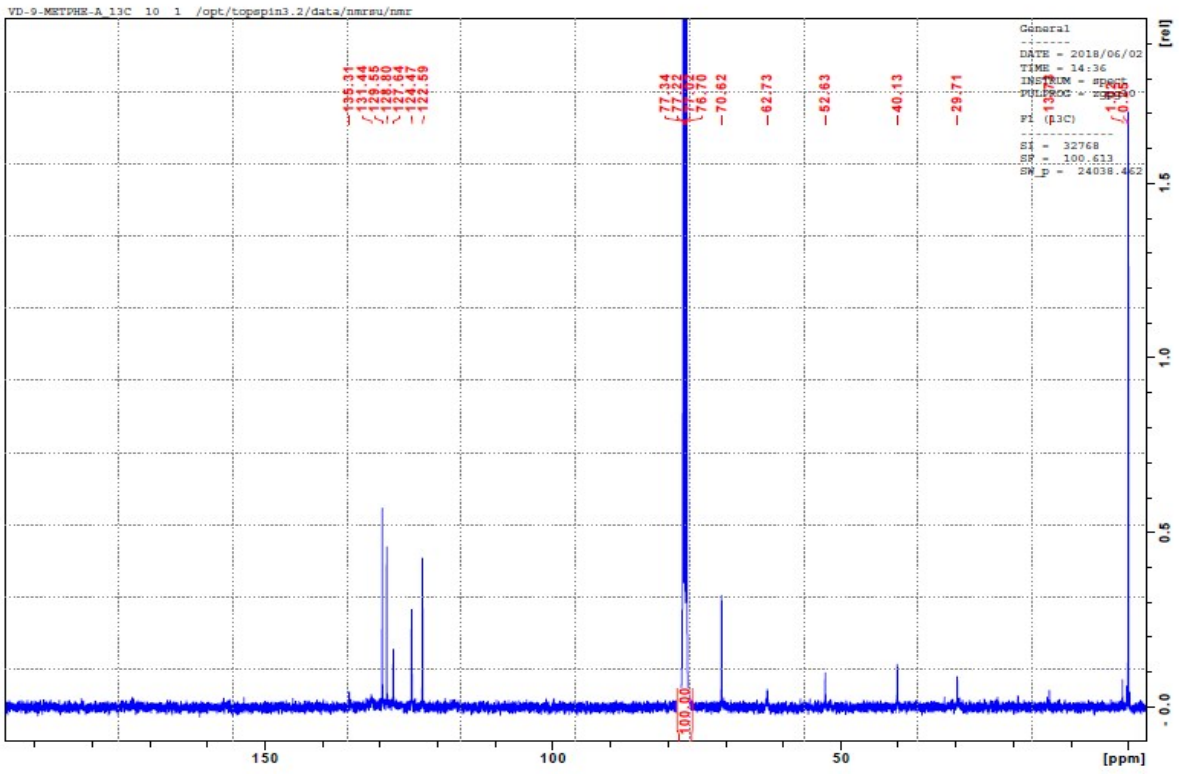


Figure S2. Compound 1 – ^{13}C NMR

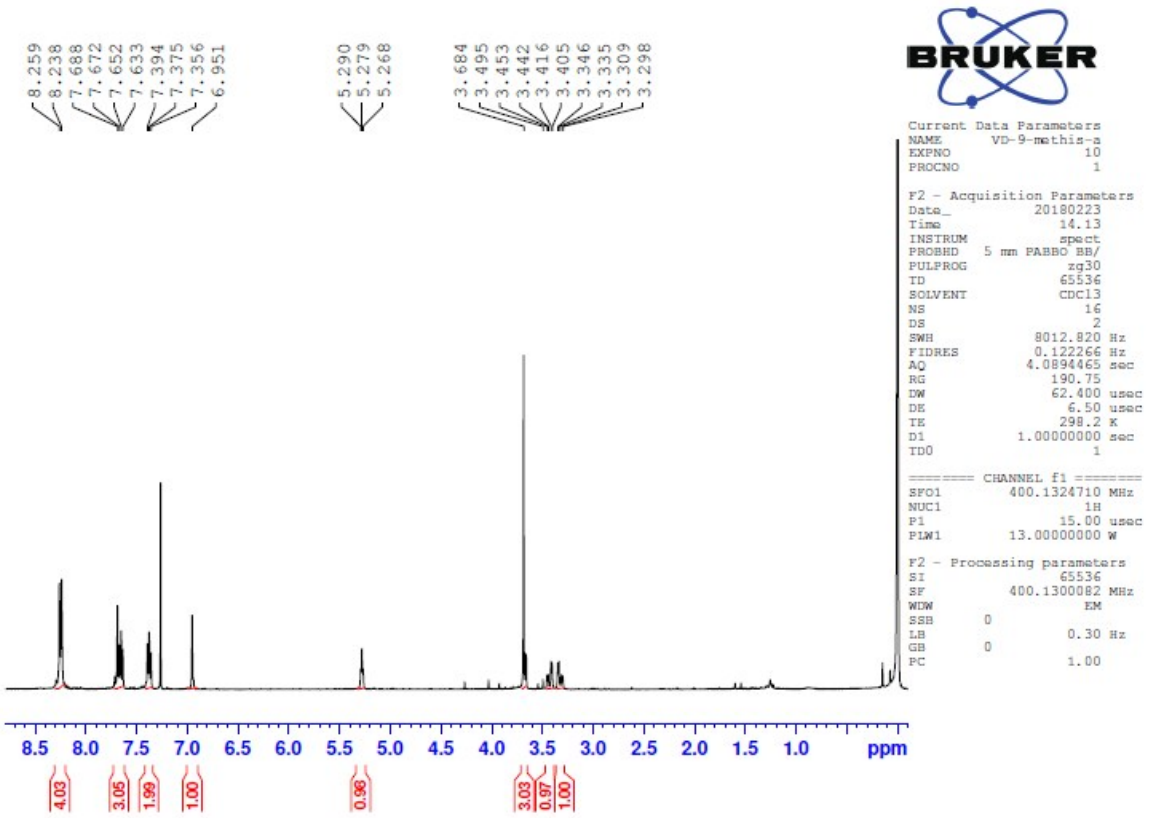


Figure S3. Compound 2 – ^{1}H NMR

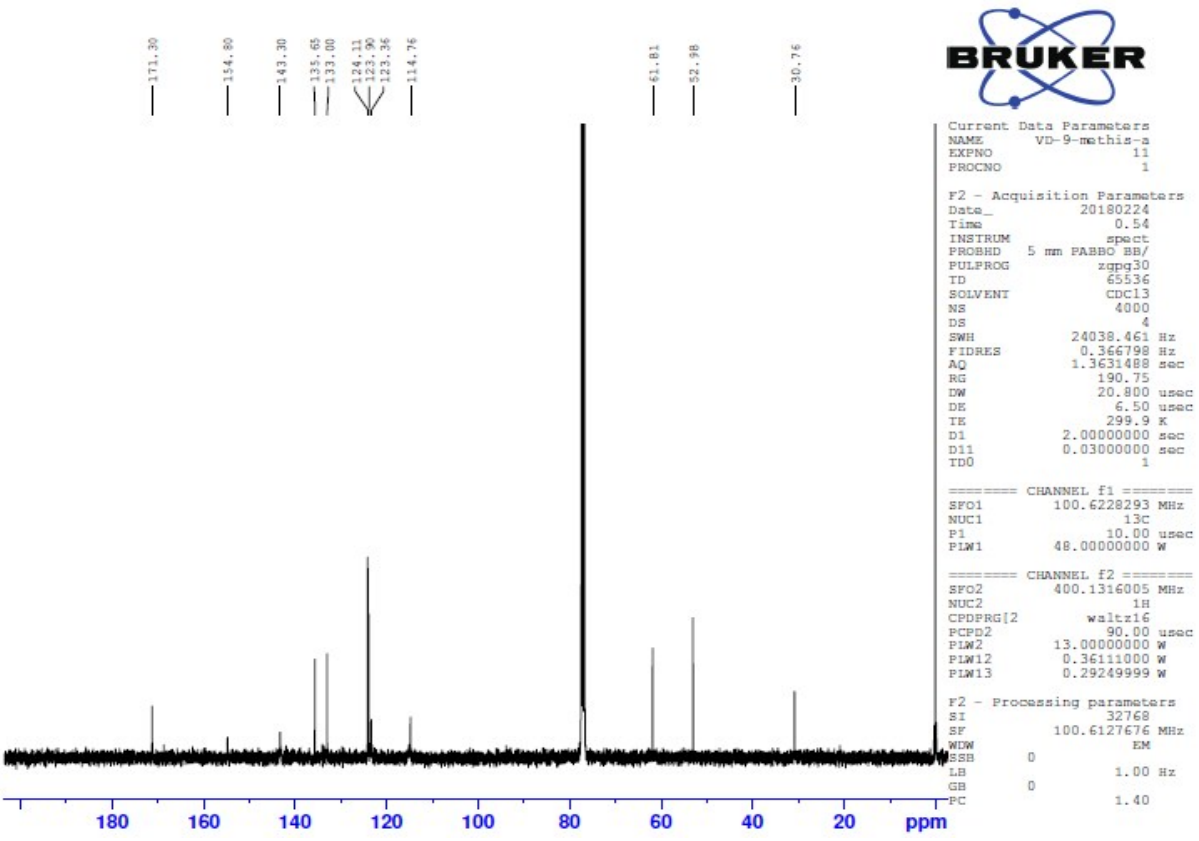


Figure S4. Compound 2 – ^{13}C NMR

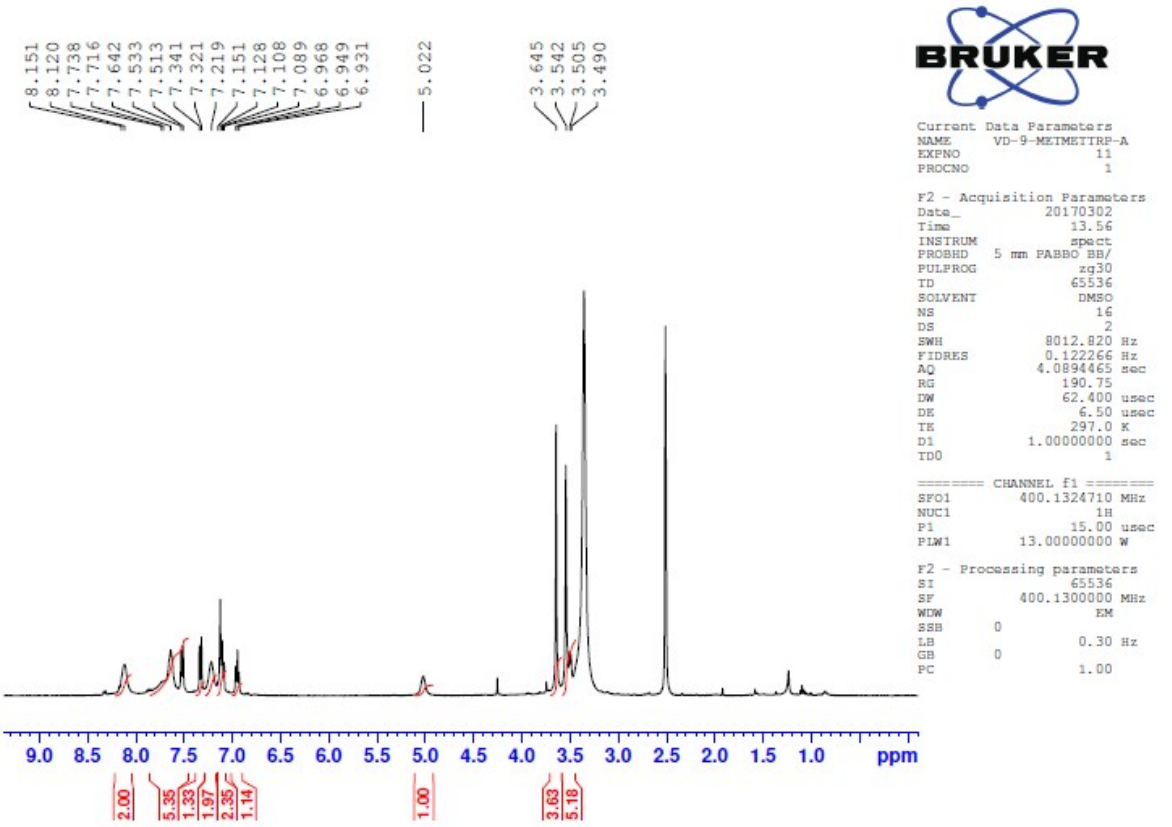


Figure S5. Compound 3 – ^{1}H NMR

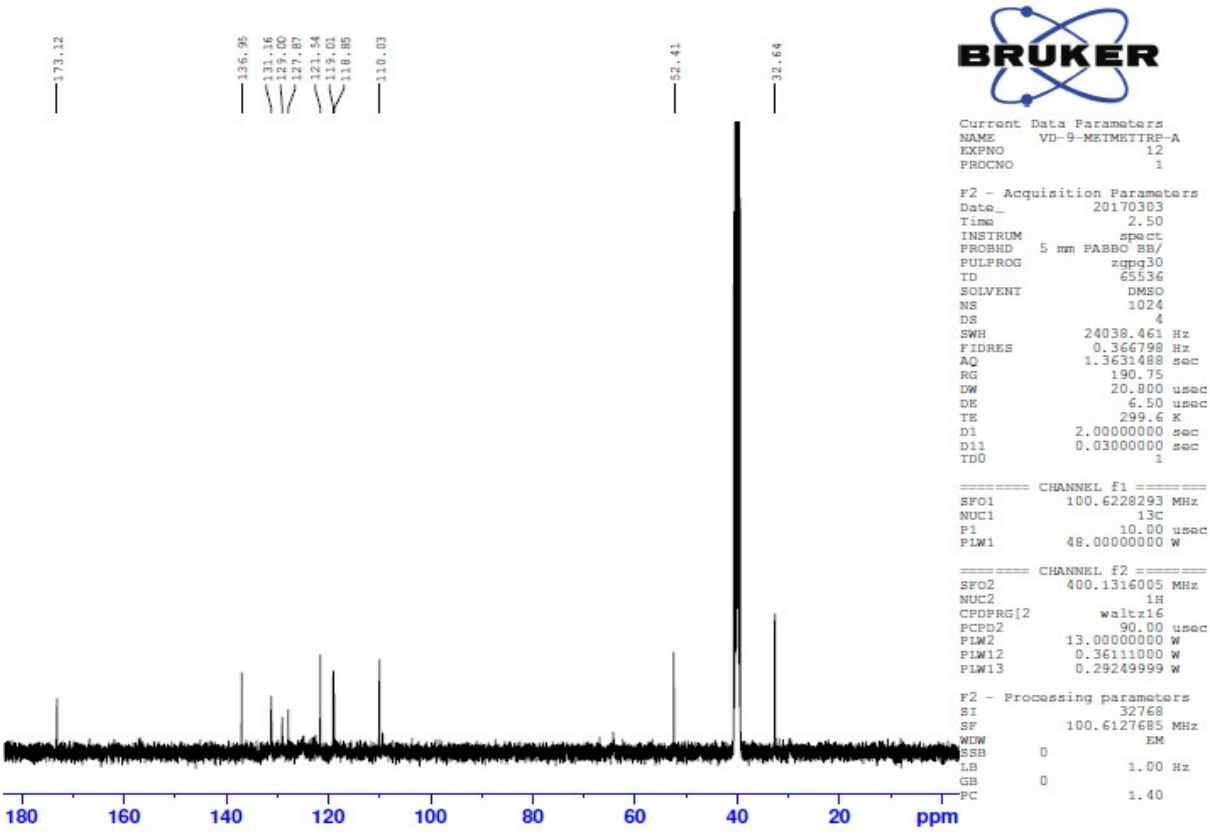


Figure S6. Compound 3 – ^{13}C NMR

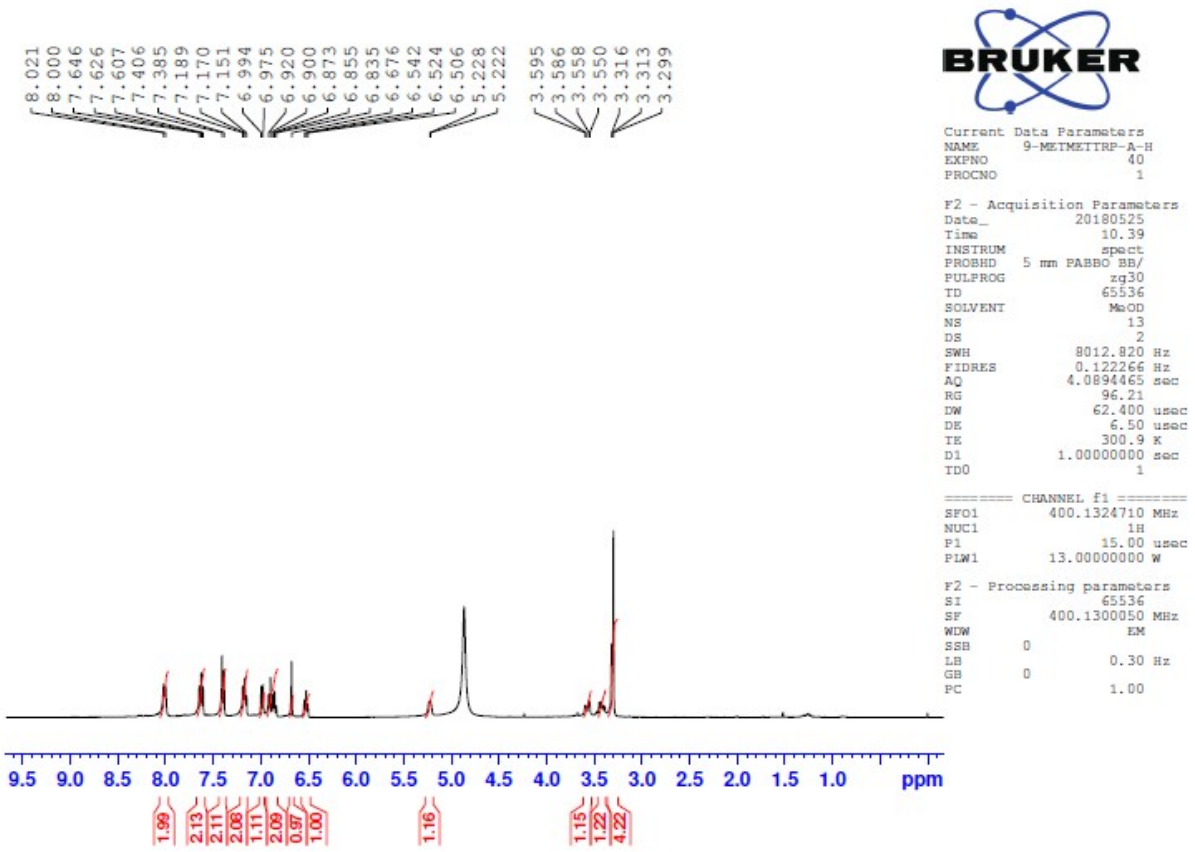


Figure S7. Compound 4 – ^{1}H NMR

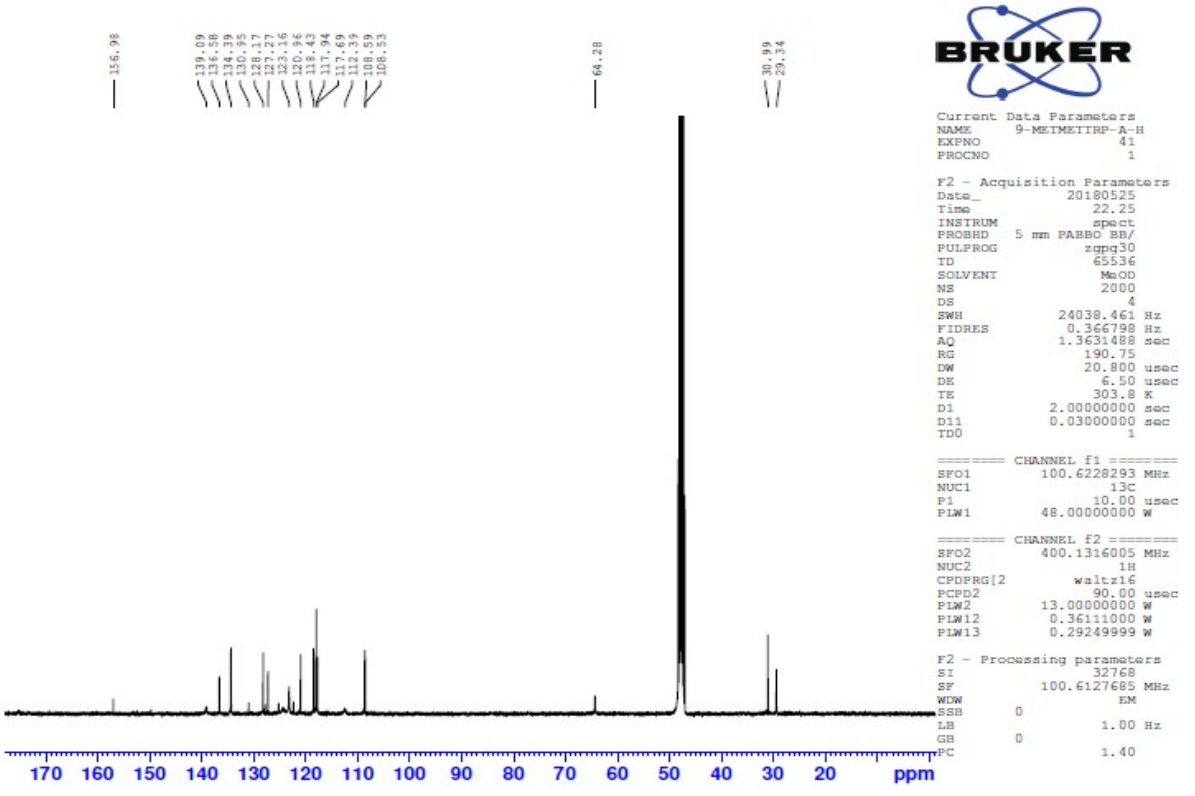


Figure S8. Compound 4 – ^{13}C NMR

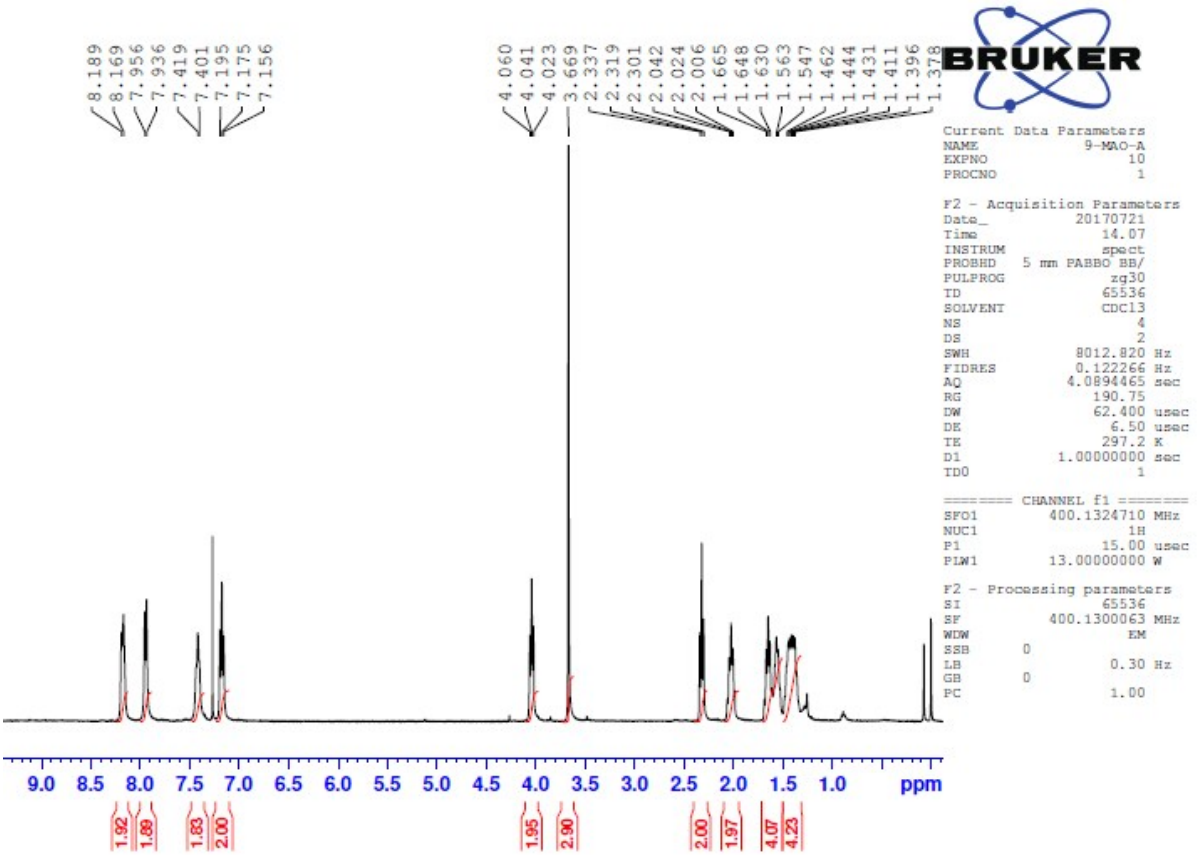


Figure S9. Compound 5 – ^{1}H NMR

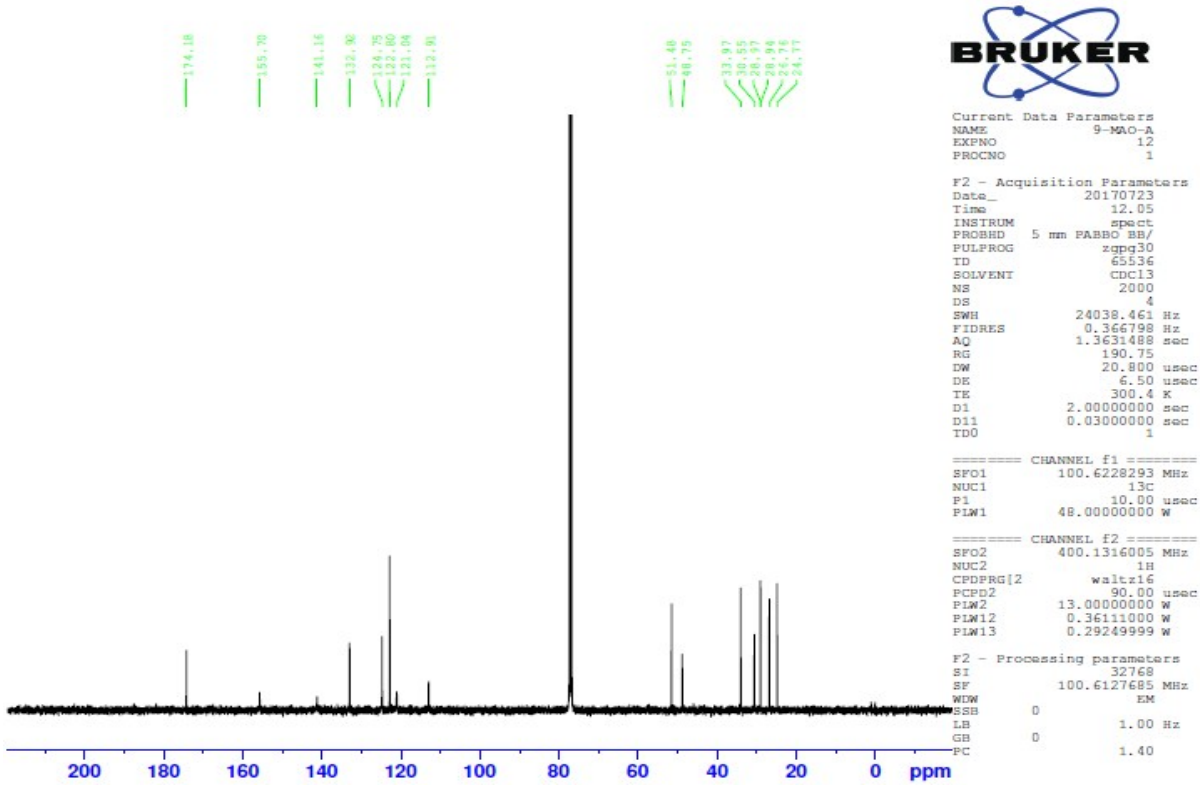


Figure S10. Compound 5 – ^{13}C NMR

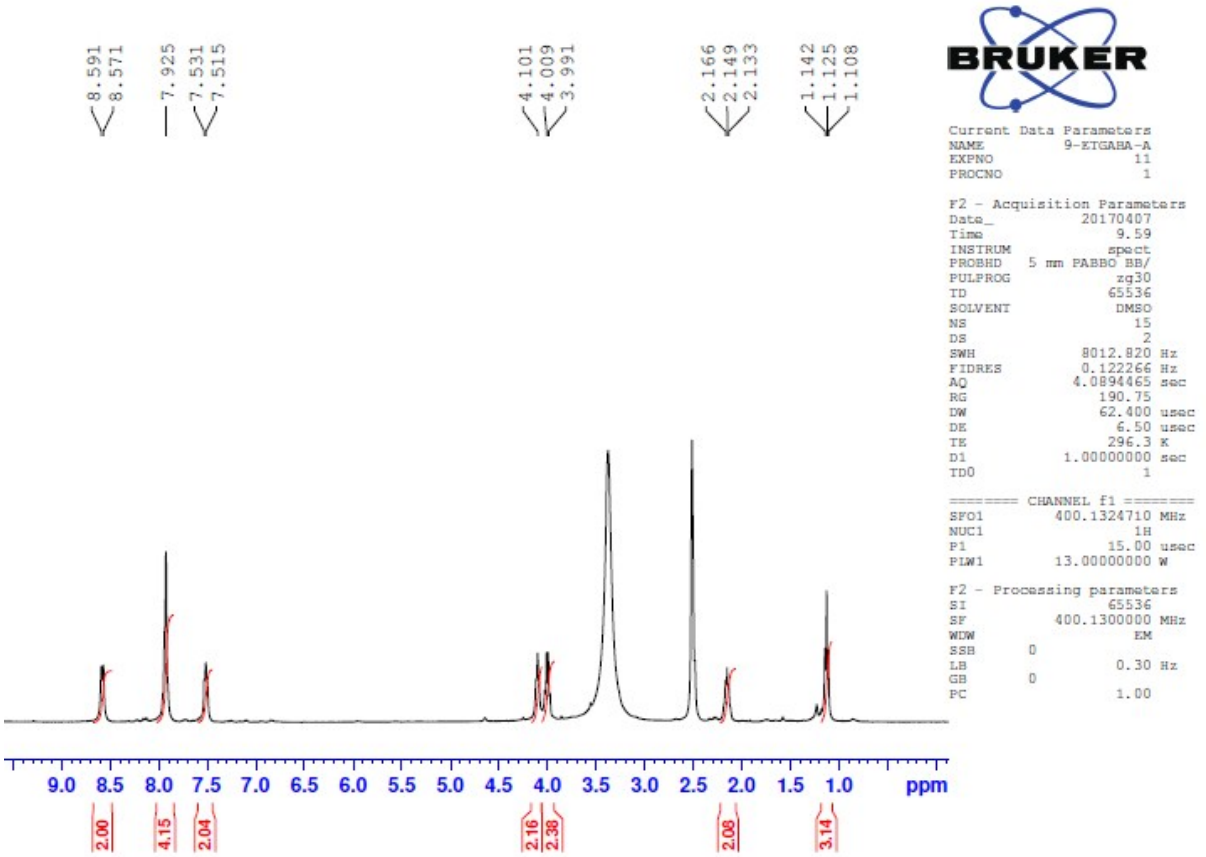


Figure S11. Compound 6 – ^{1}H NMR

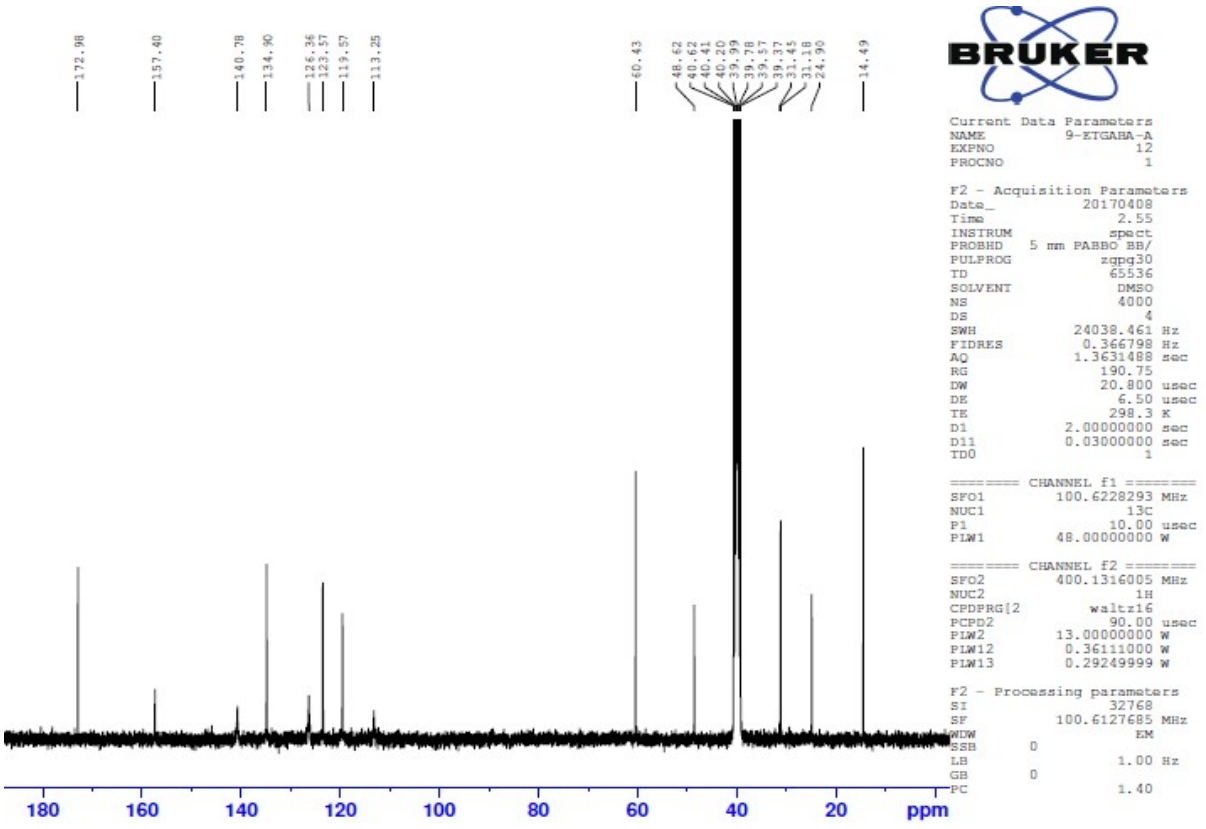
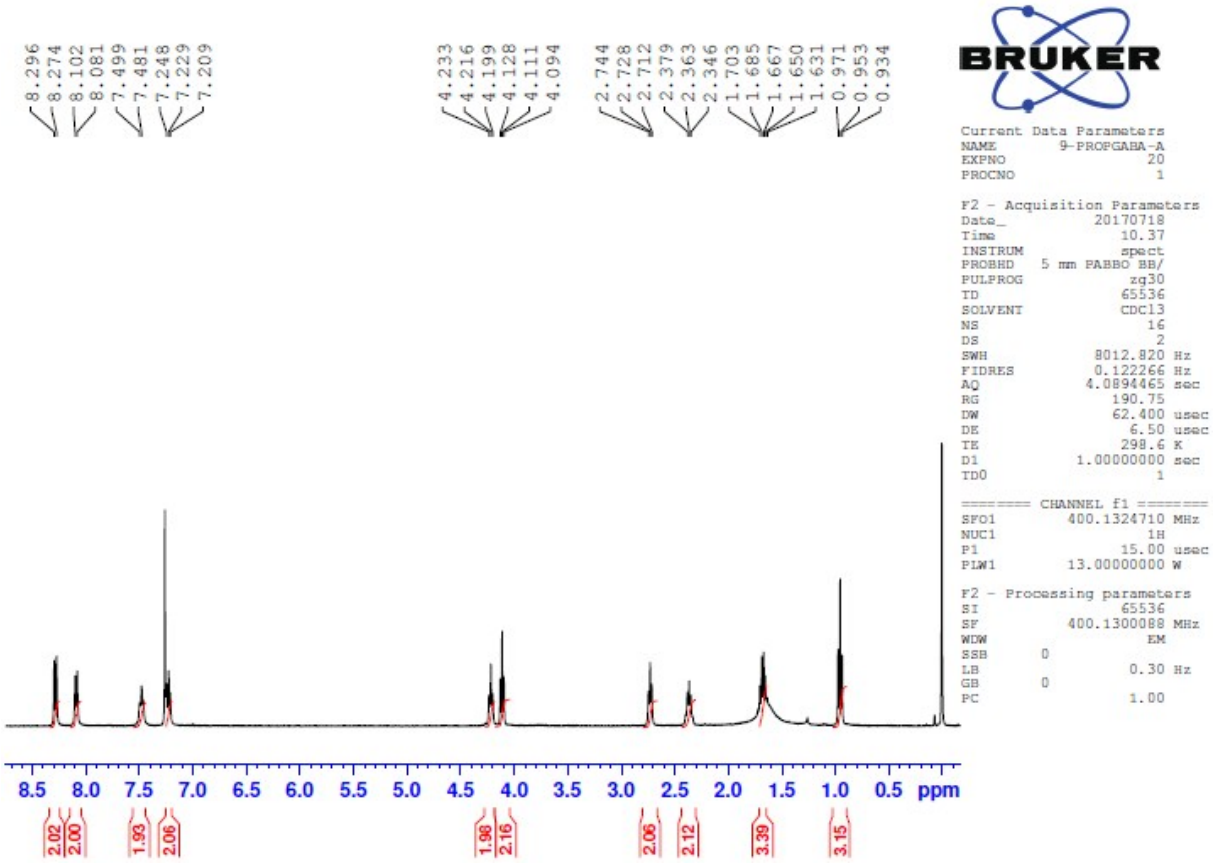


Figure S12. Compound 6 – ^{13}C NMR



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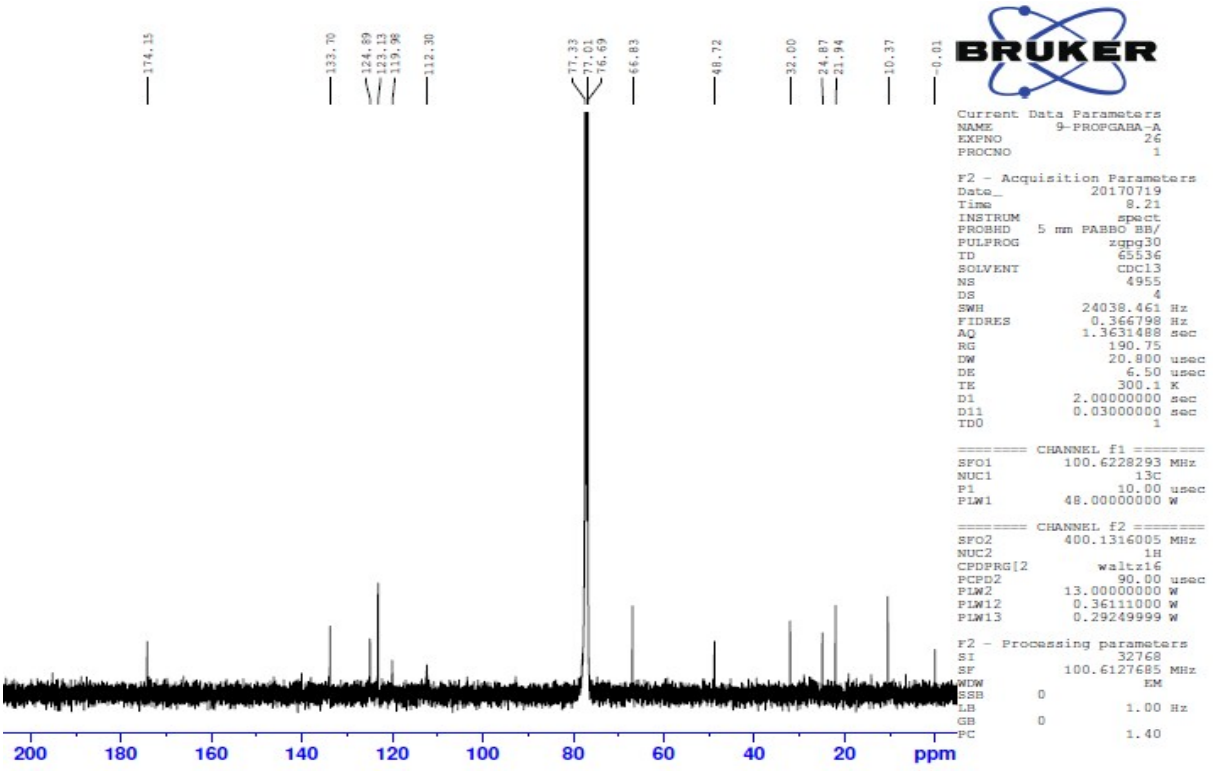

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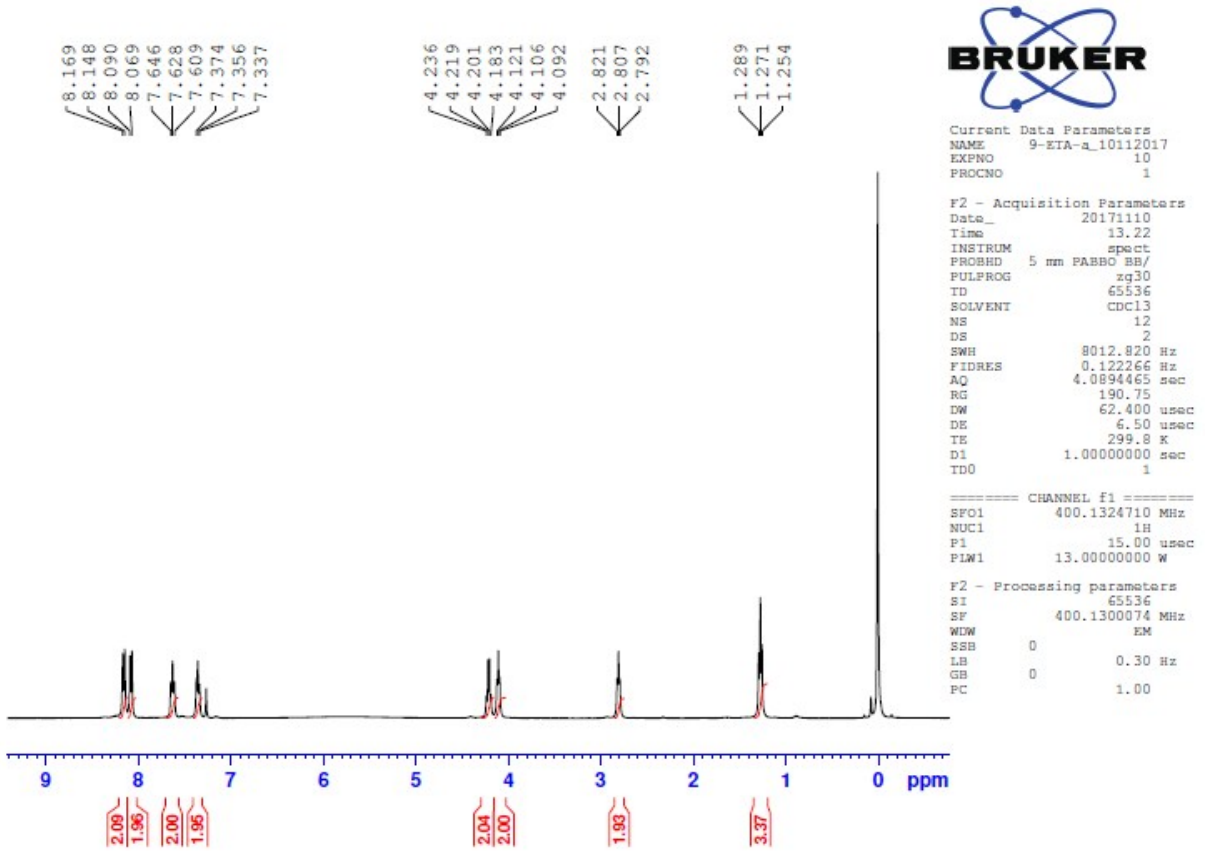

Figure S15. Compound 8 – ^{1}H NMR

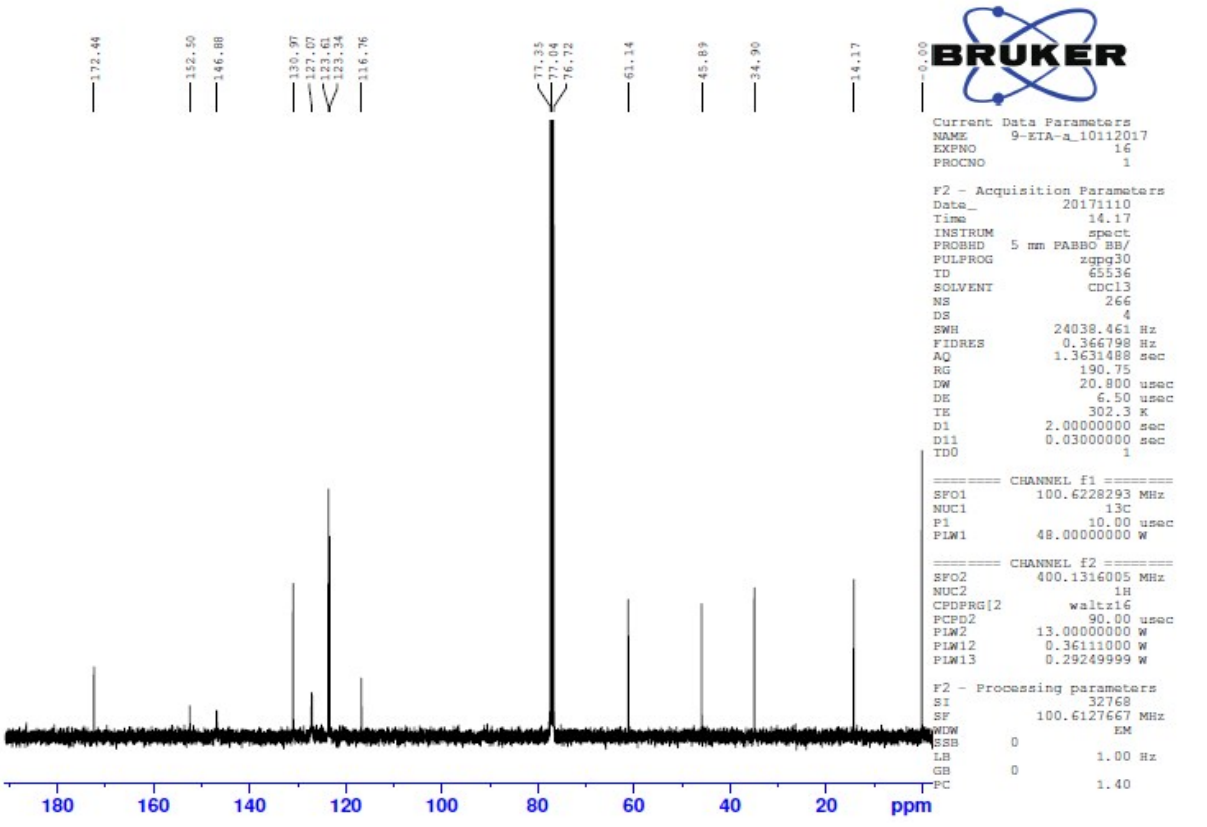

Figure S16. Compound 8 – ^{13}C NMR

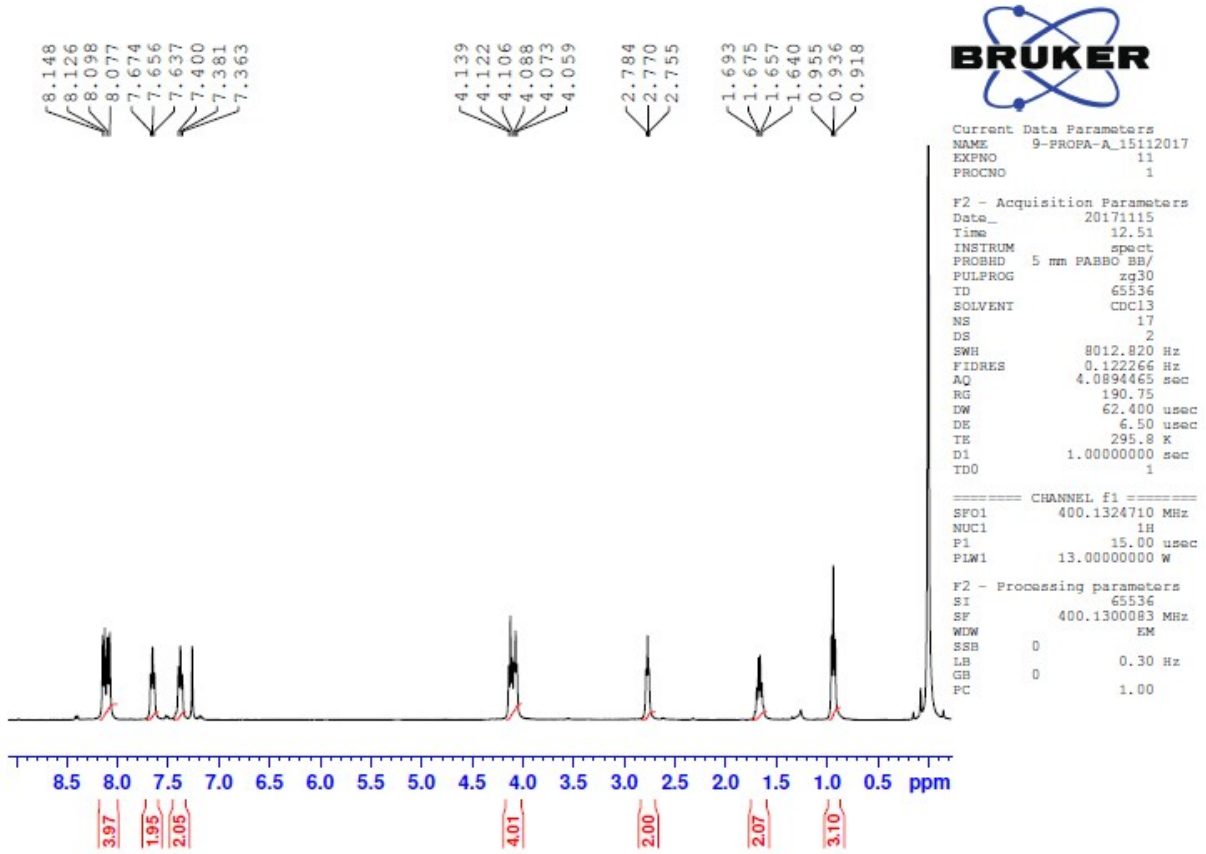

Figure S17. Compound 9 – ^{1}H NMR

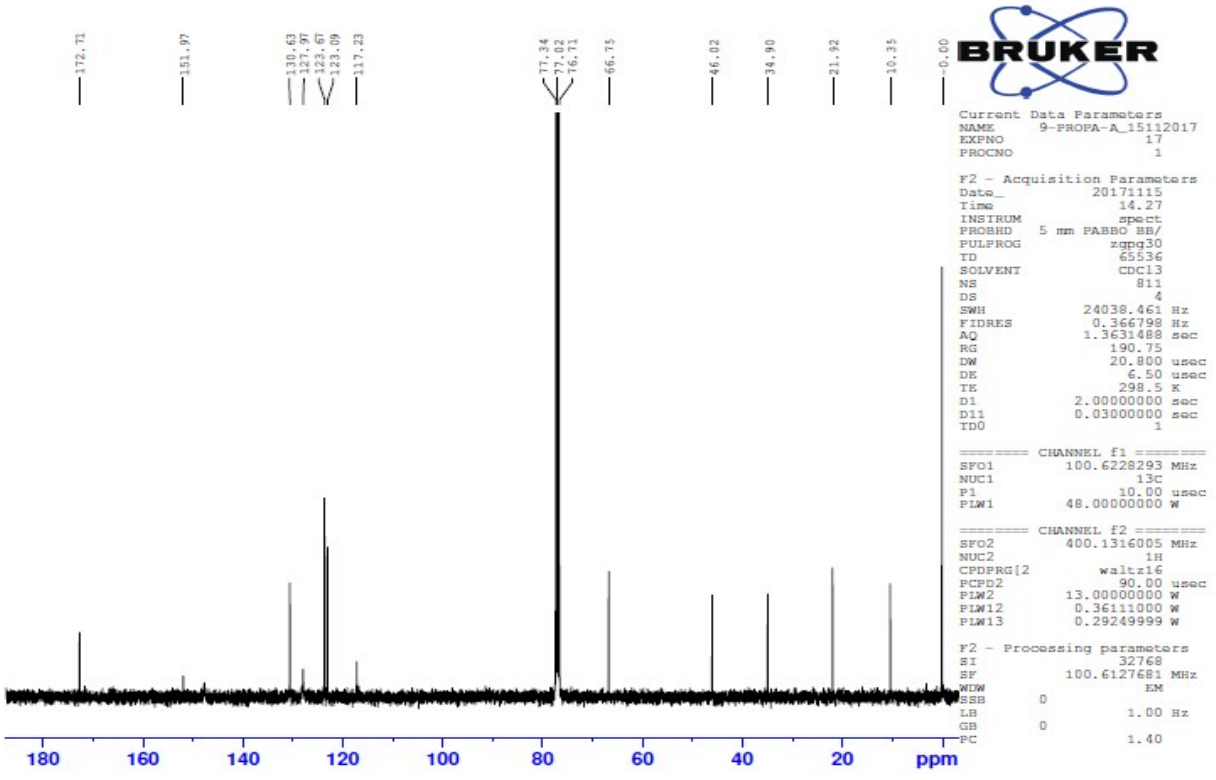

Figure S18. Compound 9 – ^{13}C NMR

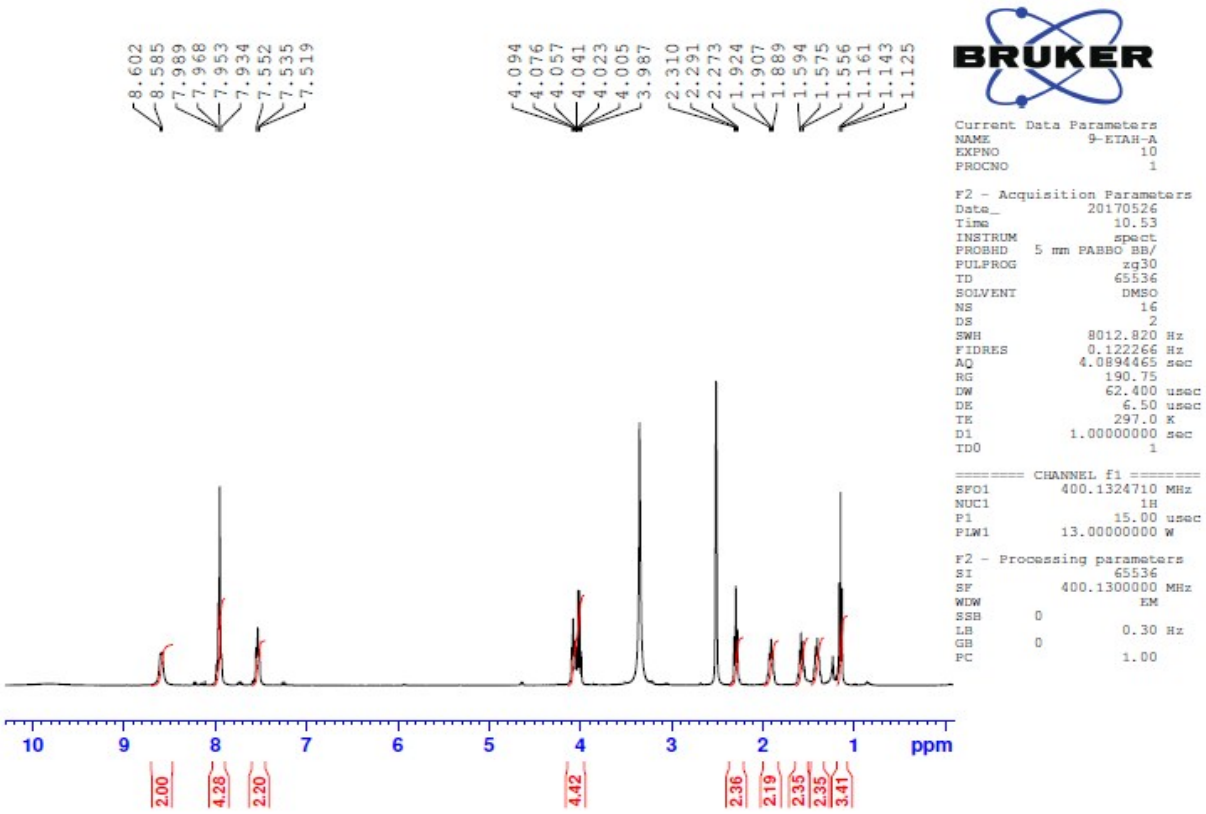

Figure S19. Compound 10 – ^{1}H NMR

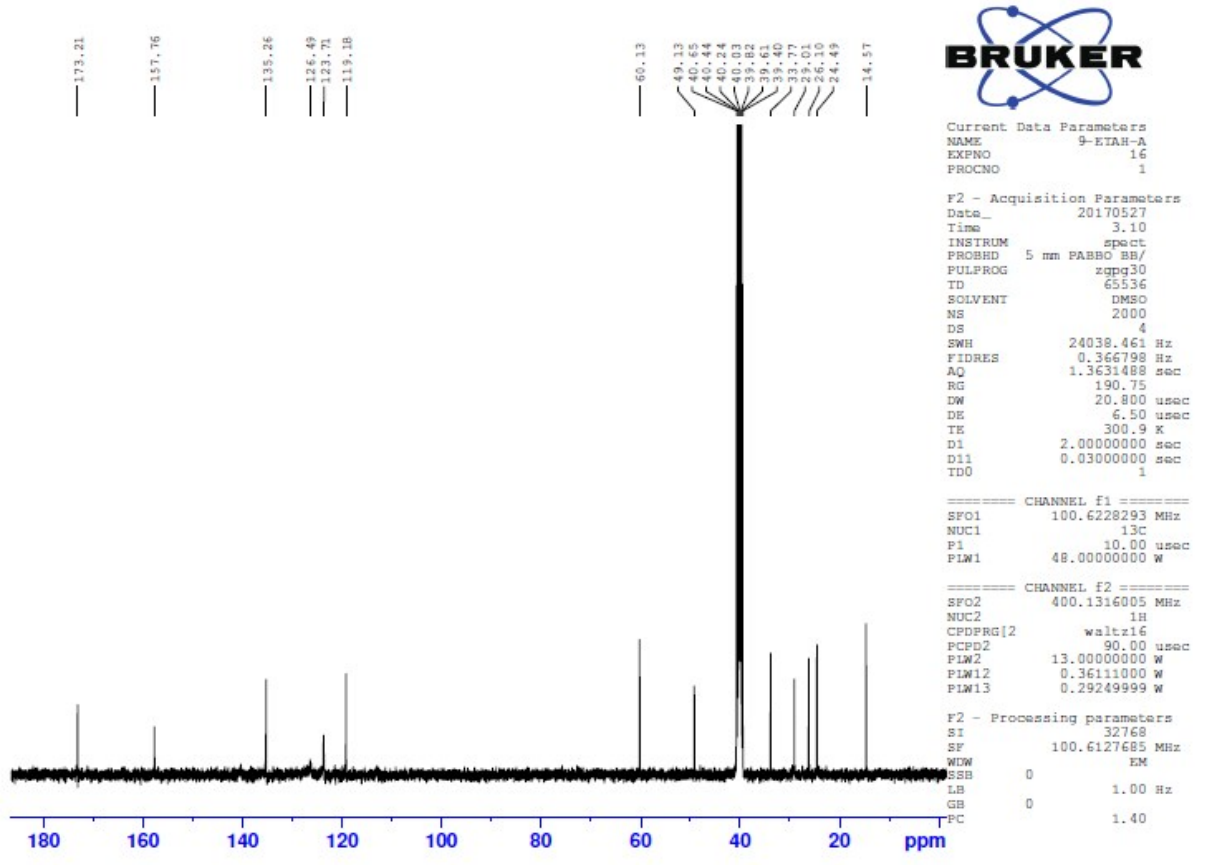

Figure S20. Compound 10 – ^{13}C NMR

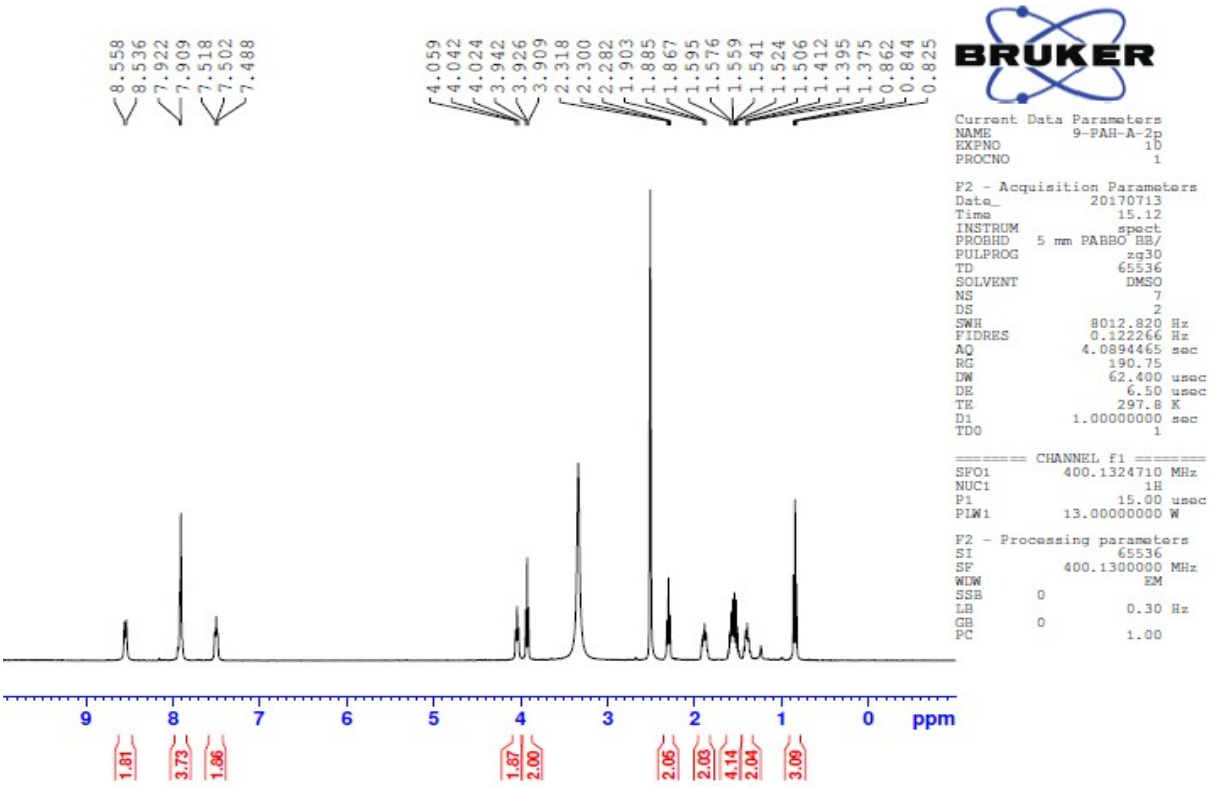

Figure S21. Compound 11 – ^{1}H NMR

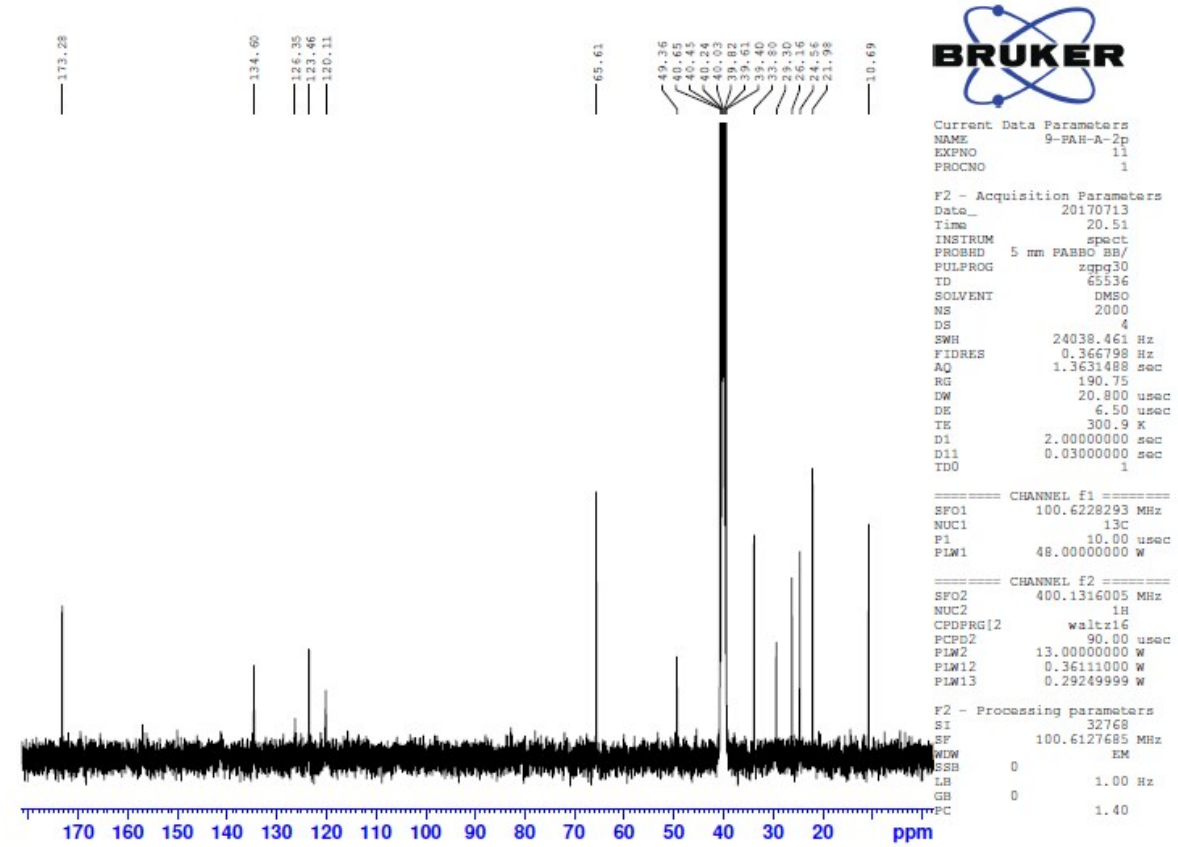

Figure S22. Compound 11 – ^{13}C NMR

HPLC analysis

The purity of tested compounds was evaluated using a HPLC method. The HPLC analysis was performed on Agilent 1200 system (Agilent Technologies, Palo Alto, CA, USA), equipped with binary pump, manual injector (20 µl sample loop) and DAD detector. The column chosen was Zorbax Extend C18 (150 mm × 4.6 mm, 5 µm particle size). The mobile phase consisted of methanol and water (pH was adjusted to 3.2 using phosphoric acid) in following ratios (v/v): 25:75 (compound **2**), 40:60 (compounds **6**, **8** and **9**), 50:50 (compound **7**), 55:45 (compound **10**) and 60:40 (compounds **1**, **3**, **4**, **5** and **11**). The column temperature was adjusted to 25ºC and the flow rate was 1 ml/min. The UV detection was performed at 220, 230, 254, 265 and 280 nm. Sample chromatograms are presented below (**Figure S23** - **Figure S33**).

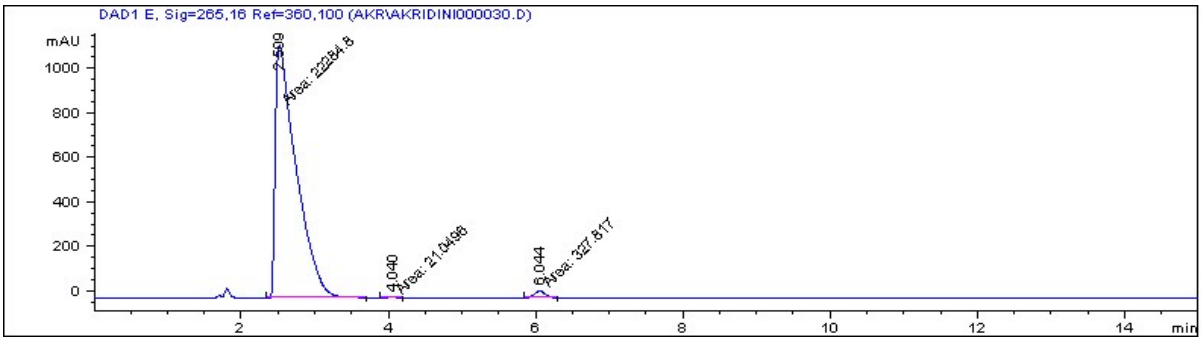

Figure S23. HPLC chromatogram - compound 1 (265 nm, purity: 98.5 %)

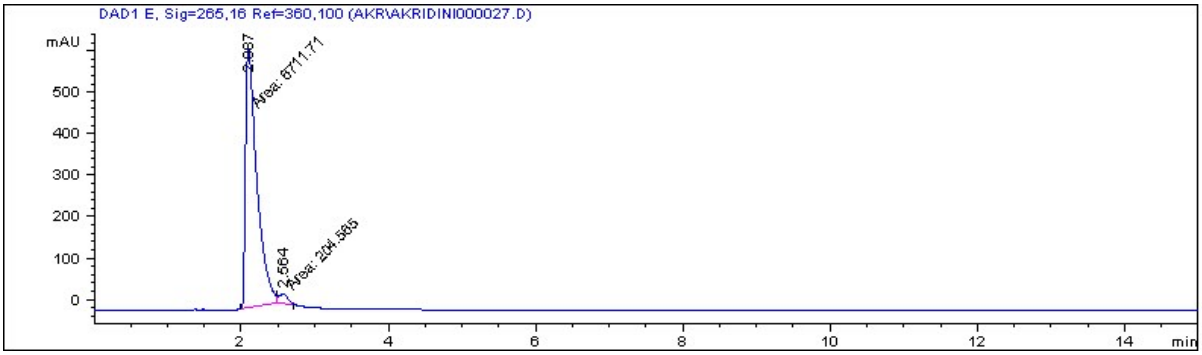

Figure S24. HPLC chromatogram - compound 2 (265 nm, purity: 97.0 %)

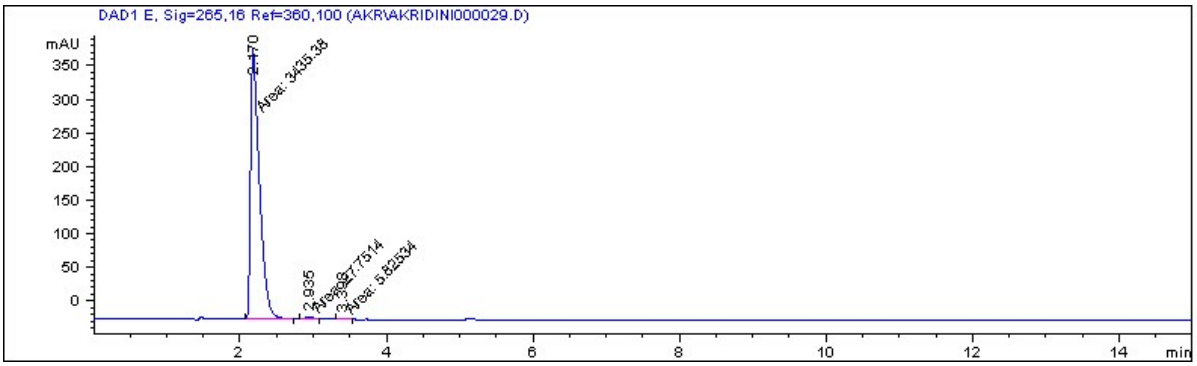

Figure S25. HPLC chromatogram - compound 3 (265 nm, purity: 99.0 %)

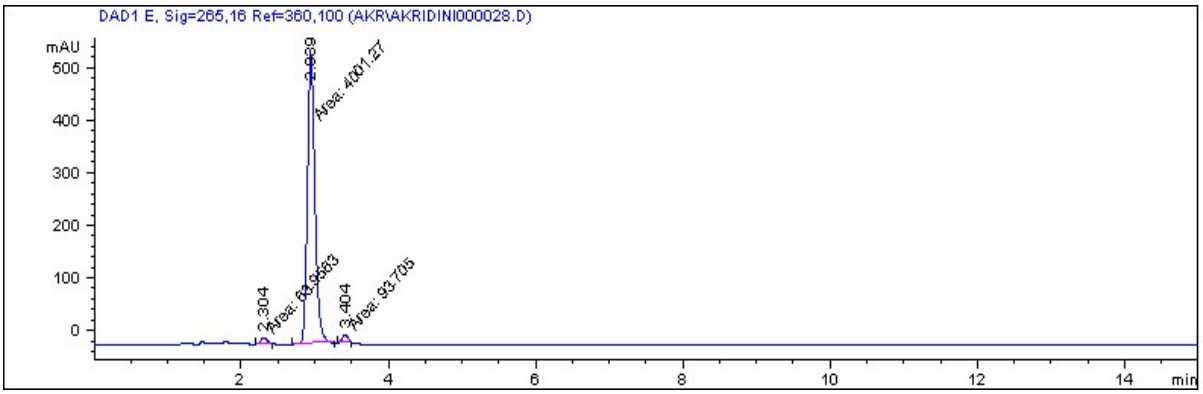

Figure S26. HPLC chromatogram - compound 4 (265 nm, purity: 96.2 %)

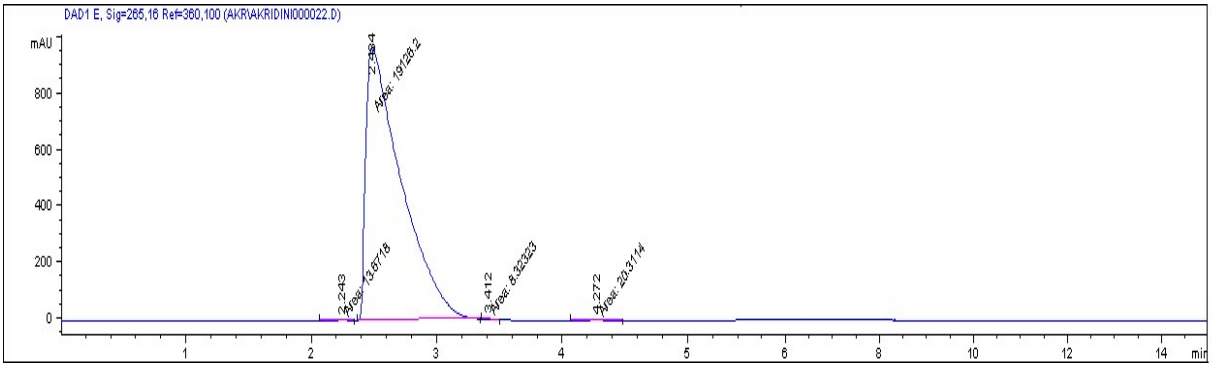

Figure S27. HPLC chromatogram - compound 5 (265 nm, purity: 99.8 %)

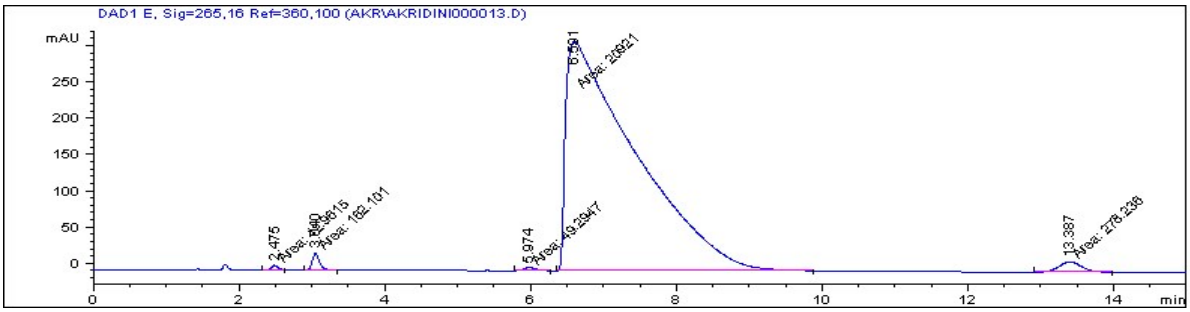

Figure S28. HPLC chromatogram - compound 6 (265 nm, purity: 97.3 %)

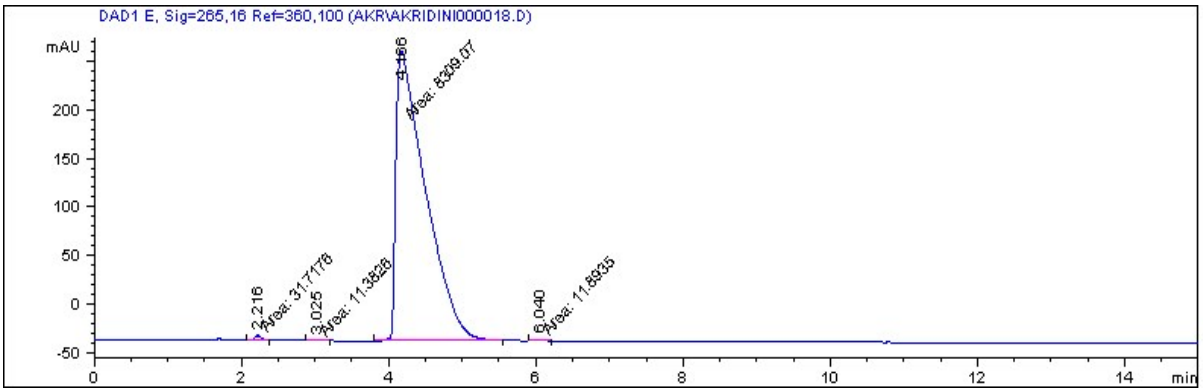

Figure S29. HPLC chromatogram - compound 7 (265 nm, purity: 99.2 %)

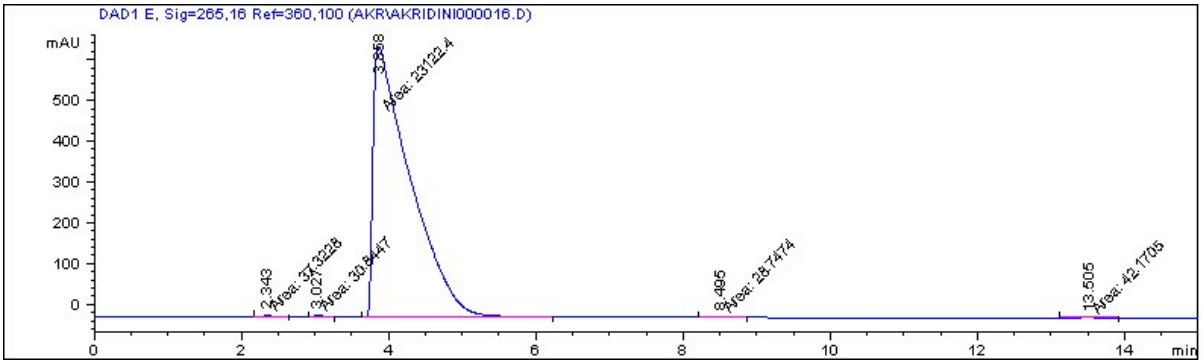

Figure S30. HPLC chromatogram - compound 8 (265 nm, purity: 99.4 %)

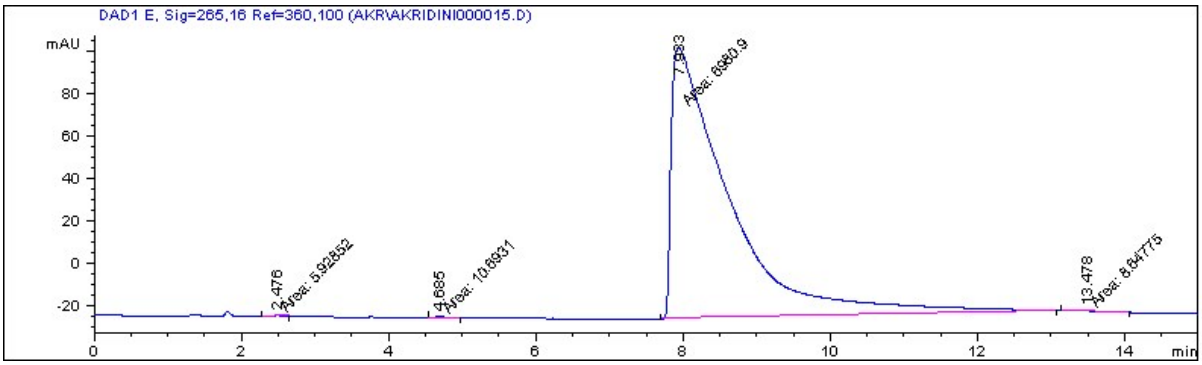

Figure S31. HPLC chromatogram - compound 9 (265 nm, purity: 99.5 %)

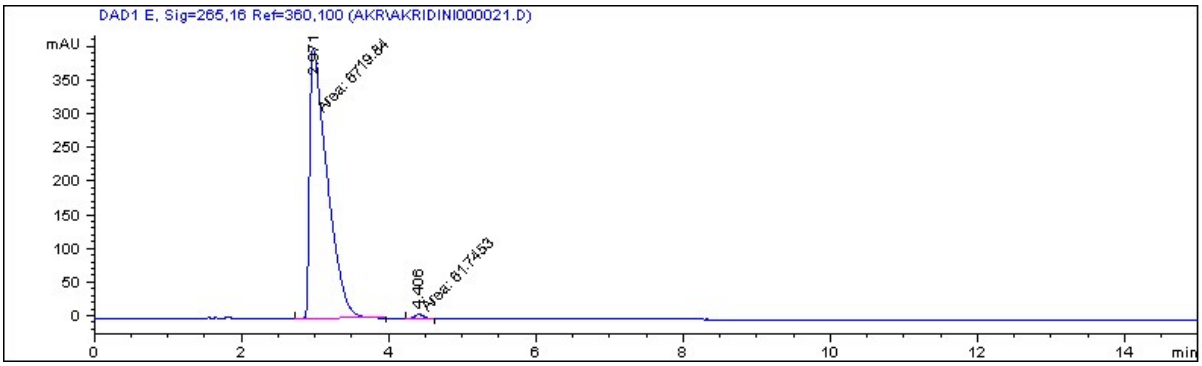

Figure S32. HPLC chromatogram - compound 10 (265 nm, purity: 99.1 %)

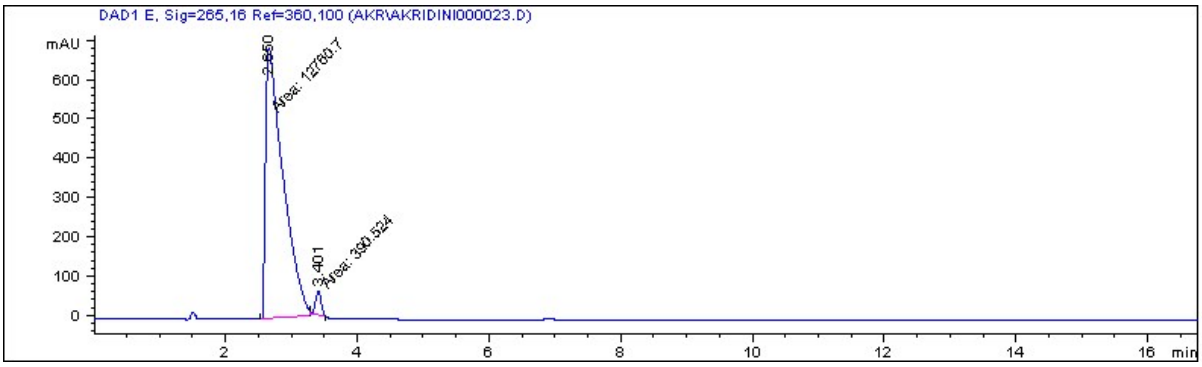

Figure S33. HPLC chromatogram - compound 11 (265 nm, purity: 97.0 %)

In silico evaluation of pharmacokinetic properties and druglikeness

Pharmacokinetic properties (absorption, distribution, metabolism and excretion - ADME) and druglikeness of derivatives **6**, **7**, **8**, **9** and amsacrine were predicted using SwissADME web service[1,2] and results are presnted in **Table S1**. Predicted pharmacokinetic properties include potential for gastrointestinal (GI) absorption, blood-brain barrier (BBB) permeability, skin permeability (permeability coefficient, LogKp), susceptibility to transport mediated by P-Glycoproteins (P-gp) and enzyme inhibition potential (CYP1A2, CYP2C19, CYP2C9, CYP2D6 and CYP3A4). Druglikeness was evaluated using Lipinski's,[3] Ghose's,[4] Veber's,[5] Egan's[6] and Muegge's[7] rules, as well as on the basis of bioavailability score.[8]

Table S1

Predicted pharmacokinetic properties and druglikeness of derivatives **6**, **7**, **8**, **9** and amsacrine.

	Compound				
Pharmacokinetic properties and druglikeness	**6**	**7**	**8**	**9**	**amsacrine**
GI absorption	High	High	High	High	High
BBB permeant	Yes	Yes	Yes	Yes	No
LogKp (skin permeation, cm/s)	-5.30	-5.01	-5.15	-4.79	-5.85
P-gp substrate	No	No	No	No	No
CYP1A2 inhibitor	Yes	Yes	Yes	Yes	Yes
CYP2C19 inhibitor	Yes	Yes	Yes	Yes	Yes
CYP2C9 inhibitor	Yes	Yes	Yes	Yes	Yes
CYP2D6 inhibitor	Yes	Yes	Yes	Yes	Yes

CYP3A4 inhibitor	Yes	Yes	Yes	Yes	Yes
Lipinski [a]	Yes	Yes	Yes	Yes	Yes
Ghose [a]	Yes	Yes	Yes	Yes	Yes
Veber [a]	Yes	Yes	Yes	Yes	Yes
Egan [a]	Yes	Yes	Yes	Yes	Yes
Muegge [a]	Yes	Yes	Yes	Yes	Yes
Bioavailability score	0.55	0.55	0.55	0.55	0.55

[a] Yes = no rule violations

All tested compounds have high predicted gastrointestinal absorption. As opposed to amsacrine, derivatives **6**, **7**, **8** and **9** were predicted to be blood-brain barrier permeants, which makes them potential candidates for treatment of brain metastases. Additionally, they were not predicted to be P-Glycoprotein substrates (P-Glycoproteins are one of the reasons for poor penetration of drugs into central nervous system,[9] which decreases their activity). Finally, these derivatives have „druglike“ structures since they obey to all imposed rules.

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1 http://www.swissadme.ch/index.php (Accessed on 25 October 2019).

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