Electronic Supplementary Material (ESI) for RSC Advances.

Supporting information

Synthesis of novel 1,5-disubstituted pyrrolo[1,2-a]quinazolines and their evaluation for anti-bacterial and anti-oxidant activities

Shaghayegh Sadat Kazemi,[a] Ali Keivanloo,*[a] Hossein Nasr-Isfahani,[a] Abdolhamid Bamoniri[b]

[a] School of Chemistry, Shahrood University of Technology, Shahrood, Iran.

[b] Chemistry Faculty, University of Kashan, Kashan, Iran.

Table of contents

General considerations

Palladium(II) chloride and propargyl alcohol were purchased from Sigma Aldrich Chemical Company. Triphenylphosphine, Anthranilic acid, Urea, *N*,*N*-dimethylaniline, triethylamine, secondary amines, thin-layer chromatography (TLC) plates, silica gel (particle size, 100-200

mesh), and all the solvents used for the reactions were purchased from Merck. NMR spectra were recorded on Bruker 400 MHz ^{1}H NMR, 300 MHz ^{1}H NMR, and 100 MHz ^{13}C NMR, 75 MHz ^{13}C NMR spectrometers. ^{1}H NMR signals were reported relative to Me_4Si (δ 0.0) or residual $CHCl_3$ (δ 7.26). ^{13}C NMR signals were reported relative to $CDCl_3$ (δ 77.16). Multiplicities were described using the following abbreviations: s = singlet, d = doublet, t = triplet and m = multiplet.IR spectra were measured on a Shimadzu IR-435 grating spectrophotometer. Mass spectra were recorded on a 5975C spectrometer manufactured in Agilent Technologies Company.

Typical procedure for preparation of quinazoline-2,4-dione

A mixture of anthranilic acid (50 g, 0.36 mol) and urea (109 g, 1.82 mol) in a 1L round bottom flask equipped with a mechanical stirrer was heated without solvent at 135-140 °C using an air condenser for 3h. The melted reaction mixture was poured into crushed ice (500 ml) with continuous stirring for 30 min. The solid so formed was filtered through Buchner funnel, washed with water (3×100 mL) and dried under vacuum over P_2O_5. The product was pure enough to use as such for next step. Yield 74%; mp >250°C.[1]

Typical procedure for preparation of 2,4-dichloroquinazoline

A mixture of quinazoline-2,4-diones (20 g, 0.12 mol), obtained above and $POCl_3$ (98 g, 0.64 mol) was refluxed in presence of N,N-dimethylaniline (8.5 g, 0.07 mol) for 5h. Reaction mixture was allowed to cool to room temperature and poured cautiously into crushed ice (500 ml) with continuous stirring for 30 min. The solid obtained was filtered through Buchner funnel, washed

with chilled alcohol (2×100 mL) and purified by column chromatography using 10% ethyl acetate/hexane as eluent. Yield 73%; mp 115- 116 °C [lit. 116-117 °C].[1]

Typical procedure for preparation of 2-chloro-4-aminoquinazoline

A mixture of 2,4-dichloroquinazoline (1 mmol, 0.194 g), secondary amines (2 mmol) in acetonitrile was refluxed for 5 hrs. until complete consumption of the starting materials monitored by TLC. After evaporation of the solvent, the resulting precipitate was washed with H_2O and did not require any further purification.[2]

Typical procedure for preparation of 2-chloro-4-alkoxyquinazoline

A mixture of sodium (1 mmol, 0.023 g) and alcohol (3 ml) was stirred 15 min at room temperature, Then, 2,4-dichloroquinazoline (1 mmol, 0.194 g) was added into the mixture until complete consumption of the starting materials monitored by TLC. After evaporation of the solvent, the resulting precipitate was washed with H_2O and did not require any further purification.[3]

Typical experimental procedure for synthesis of 1,5- disubstituted pyrrolo[1,2-a] quinazoline

A mixture of 4-substituted-2-chloroquinoxaline 2 (1 mmol), a secondary amine (3 mmol), $Pd(Ph_3P)_2Cl_2$ (0.05 mmol, 0.03 g), CuI (0.1 mmol, 0.03 g), Et_3N (4 mmol, 0.4 g) was stirred in CH_3CN (5 mL) at room temperature under argon atmosphere. Propargyl alcohol (1.2 mmol, 0.07 g) was added, and the resulting mixture was stirred at 80 °C for 15 h. After completion of the reaction, the mixture was filtered, and the remaining solid was washed with H_2O, and dried. The

crude product was purified by column chromatography (silica-gel 100) using $CHCl_3$–CH_3OH (99:1) as eluent.

5-methoxy-1-(morpholin-4-yl)pyrrolo[1,2-*a*]quinazoline (4a)

Dark Yellow solid; MP, 145-147 °C; ^{1}H NMR (300 MHz, $CDCl_3$): δ 2.45-2.59 (m, 2H, NCH_2), 3.25-3.39 (m, 2H, NCH_2), 3.84-4.05 (m, 7H, $2OCH_2$, OCH_3), 6.24 (d, *J* = 3.9 Hz, 1H, CH of pyrrole), 6.64 (d, *J* = 3.9 Hz, 1H, CH of pyrrole), 7.15-7.38 (m, 2H, Ar-H), 7.65-7.73 (m, 1H, Ar-H), 8.96 (d, *J* = 8.0 Hz, 1H, 9-H); ^{13}C NMR (75 MHz, $CDCl_3$): δ 52.45, 66.35, 67.27, 100.21, 104.34, 115.27, 122.63, 125.60, 125.88, 127.81, 134.87, 152.54, 163.65, 175.36; IR (KBr): 2940, 2830, 1610, 1500, 1120 cm^{-1}; m/z $[M]^+$ 283; HRMS for $C_{16}H_{17}N_3O_2$ calculated [MH] 283.1321; found m/z=283.1323.

5-methoxy-1-(piperidin-1-yl)pyrrolo[1,2-a]quinazoline (4b)

Orange solid; MP, 139-141 °C; ^{1}H NMR (300 MHz, $CDCl_3$): δ 1.51-1.91 (m, 6H, $3CH_2$), 2.51-2.58 (m, 2H, NCH_2), 3.21-3.43 (m, 2H, NCH_2), 4.02 (s, 3H, OCH_3), 6.21 (d, *J* = 3.9 Hz, 1H, CH of pyrrole), 6.63 (d, *J* = 4.1 Hz, 1H, CH of pyrrole), 7.04-7.21 (m, 1H, Ar-H), 7.31-7.47 (m, 1H, Ar-H), 7.56-7.64 (m, 1H, Ar-H), 8.93 (d, *J* = 8.0 Hz, 1H, 9-H); ^{13}C NMR (75 MHz, $CDCl_3$): δ 24.71, 25.15, 52.77, 67.12, 99.39, 104.53, 115.14, 121.86, 125.10, 125.48, 127.41, 135.40, 152.54, 163.75, 175.32; IR (KBr): 2944, 2830, 1620, 1505, 1125 cm^{-1}; m/z $[M]^+$ 281; HRMS for $C_{17}H_{19}N_3O$ calculated [MH] 281.1528; found m/z=281.1532.

5-ethoxy-1-(piperidin-1-yl)pyrrolo[1,2-a]quinazoline (4c)

Orange solid; MP, 139-141 °C; ^{1}H NMR (300 MHz, $CDCl_3$): δ 1.33 (t, *J* = 7.2 Hz, 3H, CH_3), 1.60-1.80 (m, 6H, $3CH_2$), 2.47-2.54 (m, 2H, NCH_2), 3.18-3.22 (m, 2H, NCH_2), 4.44 (q, *J* = 7.1

Hz, 2H, OCH_2), 6.17 (d, *J* = 4.2 Hz, 1H, CH of pyrrole), 6.58 (d, *J* = 4.2 Hz, 1H, CH of pyrrple), 7.10-7.19 (m, 1H, Ar-H), 7.21-7.32 (m, 1H, Ar-H), 7.53-7.61 (m, 1H, Ar-H), 8.91 (d, *J* = 9.0 Hz, 1H, 9-H); ^{13}C NMR (75 MHz, $CDCl_3$): δ 14.43, 24.87, 25.94, 53.85, 62.36, 100.39, 105.63, 116.21, 122.86, 126.11, 126.76, 127.75, 136.50, 154.22, 162.51, 176.62; IR (KBr): 2950, 2850, 1615, 1510, 1120 cm^{-1}; m/z $[M]^+$ 295; HRMS for $C_{18}H_{21}N_3O$ calculated [MH] 295.1685; found m/z=295.1686.

1-(morpholin-4-yl)-5-propoxypyrrolo[1,2-*a*]quinazoline (4d)

Orange solid; MP, 127-129 °C; ^1H NMR (300 MHz, $CDCl_3$): δ 0.81 (t, *J* = 7.3 Hz, 3H, CH_3), 1.58-1.77 (m, 2H, CH_2), 2.44-2.58 (m, 2H, NCH_2), 3.29-3.32 (m, 2H, NCH_2), 3.74-3.92 (m, 4H, $2OCH_2$), 4.24 (t, *J* = 6.6 Hz, 2H, OCH_2), 6.18 (d, *J* = 4.2 Hz, 1H, CH of pyrrole), 6.59 (d, *J* = 4.2 Hz, 1H, CH of pyrrole); 7.23-7.47 (m, 2H, Ar-H), 7.53-7.65 (m, 1H, Ar-H), 8.93 (d, *J* = 8.1 Hz, 1H, 9-H); ^{13}C NMR (75 MHz, $CDCl_3$): δ 10.90, 14.76, 52.03, 66.19, 68.26, 100.36, 104.22, 117.89, 121.00, 126.12, 126.42, 128.90, 136.76, 154.19, 163.37, 175.42; IR (KBr): 2950, 2840, 1620, 1500, 1110 cm^{-1}; m/z $[M]^+$ 311; HRMS for $C_{18}H_{21}N_3O_2$ calculated [MH] 311.1634; found m/z=311.1629.

1-(piperidin-1-yl)-5-propoxypyrrolo[1,2-*a*]quinazoline (4e)

Brown solid; MP, 125-126 °C; ^1H NMR (300 MHz, $CDCl_3$): δ 0.82 (t, *J* = 7.3 Hz, 3H, CH_3), 1.18-1.37 (m, 2H, CH_2), 1.57-1.71 (m, 6H, $3CH_2$), 2.51-2.59 (m, 2H, NCH_2), 3.22-3.25 (m, 2H, NCH_2), 4.14 (t, *J* = 6.6 Hz, 2H, OCH_2), 6.21 (d, *J* = 4.2 Hz, 1H, CH of pyrrole), 6.63 (d, *J* = 4.2 Hz, 1H, CH of pyrrole), 7.15-7.18 (m, 1H, Ar-H), 7.43-7.47 (m, 1H, Ar-H), 7.55-7.63 (m, 1H, Ar-H), 8.93 (d, *J* = 8.4 Hz, 1H, 9-H); ^{13}C NMR (75 MHz, $CDCl_3$): δ 10.98, 14.08, 23.76, 25.77,

53.83, 68.16, 100.40, 104.38, 116.19, 122.87, 126.61, 127.68, 128.81, 135.46, 154.11, 165.78, 175.28; IR (KBr): 2955, 2840, 1615, 1515, 1110 cm^{-1}; m/z $[M]^+$ 309; HRMS for $C_{19}H_{23}N_3O$ calculated [MH] 309.1841; found m/z=309.1846.

5-butoxy-1-(piperidin-1-yl)pyrrolo[1,2-a]quinazoline (4f)

Orange solid; MP, 117-119 °C; ^1H NMR (300 MHz, $CDCl_3$): δ 0.85 (t, *J* = 7.4 Hz, 3H, CH_3), 1.17-1.47 (m, 4H, $2CH_2$), 1.65-1.79 (m, 6H, $3CH_2$), 2.51-2.58 (m, 2H, NCH_2), 3.21-3.25 (m, 2H, NCH_2), 4.14 (t, *J* = 6.6 Hz, 2H, OCH_2), 6.21 (d, *J* = 4.2 Hz, 1H, CH of pyrrole), 6.62 (d, *J* = 4.0 Hz, 1H, CH of pyrrole), 7.12-7.18 (m, 1H, Ar-H), 7.44-7.56 (m, 2H, Ar-H), .8.93 (d, *J* = 7.8 Hz, 1H, 9-H); ^{13}C NMR (75 MHz, $CDCl_3$): δ 10.96, 13.94, 19.34, 24.06, 26.20, 57.34, 68.04, 100.38, 105.69, 116.20, 122.85, 126.66, 127.71, 128.82, 135.05, 154.18, 168.20, 175.18; IR (KBr): 2950, 2850, 1615, 1525, 1110 cm^{-1}; m/z $[M]^+$ 323; HRMS for $C_{20}H_{25}N_3O$ calculated [MH] 323.1998; found m/z=323.1995.

5-butoxy-1-(morpholin-4-yl)pyrrolo[1,2-*a*]quinazoline (4g)

Orange solid; MP, 121-123 °C; ^1H NMR (300 MHz, $CDCl_3$): δ 0.82 (t, *J* = 7.4 Hz, 3H, CH_3), 1.15-1.37 (m, 2H, CH_2), 1.57-1.72 (m, 2H, CH_2), 2.51-2.56 (m, 2H, NCH_2), 3.22-3.57 (m, 2H, NCH_2),), 4.14 (t, *J* = 6.6 Hz, 2H, OCH_2), 6.21 (d, *J* = 4.2 Hz, 1H, CH of pyrrole), 6.64 (d, *J* = 4.2 Hz, 1H, CH of pyrrole), 7.17-7.21 (m, 1H, Ar-H), 7.43-7.47 (m, 1H, Ar-H), 7.55-7.64 (m, 1H, Ar-H), 8.93 (d, *J* = 8.1 Hz, 1H, 9-H); ^{13}C NMR (75 MHz, $CDCl_3$): δ 10.98, 14.08, 19.38, 57.56, 66.75, 68.17, 101.84, 104.14, 116.31, 121.44, 126.43, 126.72, 128.82, 136.36, 155.01, 167.80, 175.83; IR (KBr): 2955, 2850, 1610, 1520, 1115 cm^{-1}; m/z $[M]^+$ 325; HRMS for $C_{19}H_{23}N_3O_2$ calculated [MH] 325.1790; found m/z=325.1794.

1,5-di(morpholin-4-yl)pyrrolo[1,2-*a*]quinazoline (4h):

Yellow solid; MP, 149-151 °C; ^{1}H NMR (400 MHz, $CDCl_3$): δ 2.45-2.60 (m, 2H, NCH_2), 3.16-3.18 (m, 2H, NCH_2), 3.33-3.56 (m, 4H, $2NCH_2$), 3.68-4.10 (m, 8H, $4OCH_2$,), 6.38 (d, J = 4.0 Hz, 1H, CH of pyrrole), 6.83 (d, J = 4.0 Hz, 1H, CH of pyrrole), 7.25-7.27 (m, 1H, Ar-H), 7.48-7.50 (m, 1H, Ar-H), 7.65-7.67 (m, 1H, Ar-H), 8.90 (d, J = 9.0 Hz, 1H, 9-H); ^{13}C NMR (75 MHz, $CDCl_3$): δ 52.12, 54.38, 66.25, 64.37, 100.26, 105.31, 115.23, 122.51, 125.37, 126.11, 127.48, 136.05, 152.89, 162.28, 171.23; IR (KBr): 2950, 2850, 1610, 1520, 1115 cm^{-1}; m/z $[M]^+$ 338; HRMS for $C_{19}H_{22}N_4O_2$ calculated [MH] 338.1743; found m/z=338.1745.

1-(morpholin-4-yl)-5-(piperidin-1-yl)pyrrolo[1,2-*a*]quinazoline (4i)

Yellow solid; MP, 145-147 °C; ^{1}H NMR (300 MHz, $CDCl_3$): δ 1.61-1.85 (m, 6H, $3CH_2$), 2.34-2.49 (m, 2H, NCH_2), 3.10-3.21 (m, 2H, NCH_2), 3.61-3.72 (m, 4H, $2NCH_2$), 3.96-4.09 (m, 4H, $2OCH_2$), 6.38 (d, J = 4.2 Hz, 1H, CH of pyrrole), 6.75 (d, J = 4.2 Hz, 1H, CH of pyrrole), 7.19-7.42 (m, 2H, Ar-H), 7.64-7.69 (m, 1H, Ar-H), 8.86 (d, J = 8.0 Hz, 1H, 9-H); ^{13}C NMR (75 MHz, $CDCl_3$): δ 24.84, 25.94, 51.20, 53.37, 66.42, 100.39, 104.83, 115.42, 122.74, 125.53, 126.12, 135.84, 153.07, 161.27, 171.13; IR (KBr): 2950, 2850, 1600, 1510, 1100 cm^{-1}; m/z $[M]^+$ 336; HRMS for $C_{20}H_{24}N_4O$ calculated [MH] 336.1950; found m/z=336.1951.

Anti-bacterial assay

The anti-bacterial activities of pyrrolo[1,2-a]quinazolines were evaluated biologically using a well-diffusion method. First the nutrient agar and nutrient broth cultures were prepared according to the manufacturer's instructions, and they were then incubated at 37 °C. After

incubation for the appropriate time period, a suspension of 30 μL of each bacterium was added to the nutrient agar plates. Cups (5 mm in diameter) were cut in the agar using a sterilized glass tube. Each well received 30 μL of the test compounds at a concentration of 1000 μg/ml in DMSO. Then the plates were incubated at 37 °C for 24 h, after which time, the inhibition zone was measured. The values were expressed in millimeters (mm). The anti-bacterial activity of each pyrrolo[1,2-a]quinazoline was compared with that for PenicillinGand as the standard. DMSO was used as the negative control.

DPPH radical scavenging assay

The DPPH radical scavenging activities of **4a**, **4d**, **4e**, **4f**, and **4h** were evaluated according to the literature.[23] The DPPH solution was prepared by dissolving an appropriate amount of DPPH in MeOH to give a concentration of 6.25×10^{-5} M. Compounds **4a**, **4d**, **4e**, **4f**, **4h**, and DPPH with different concentrations (4000, 2000, 1000, 500, 250, and 125 μg/mL) in MeOH were prepared. Then 0.1 mL of each pyrrolo[1,2-a]quinazoline solution was added to 3.9 mL of the DPPH solution, and was shaken vigorously. The samples were kept in dark for 30 min, and then their absorbance was measured at 517 nm. MeOH was used as the blank. The radical scavenging activity was calculated as follows:

$$\text{Radical scavenging activity (\%)} = \left(\frac{A_{Control} - A_{Sample}}{A_{Control}}\right) * 100\%$$

where $A_{control}$ is the absorbance of the negative control (containing all reagents except the test compounds) and A_{sample} is the absorbance of the test compounds. IC_{50} values of the test compounds were determined by plotting the radical scavenging activity percentage against the concentration of the test compound.

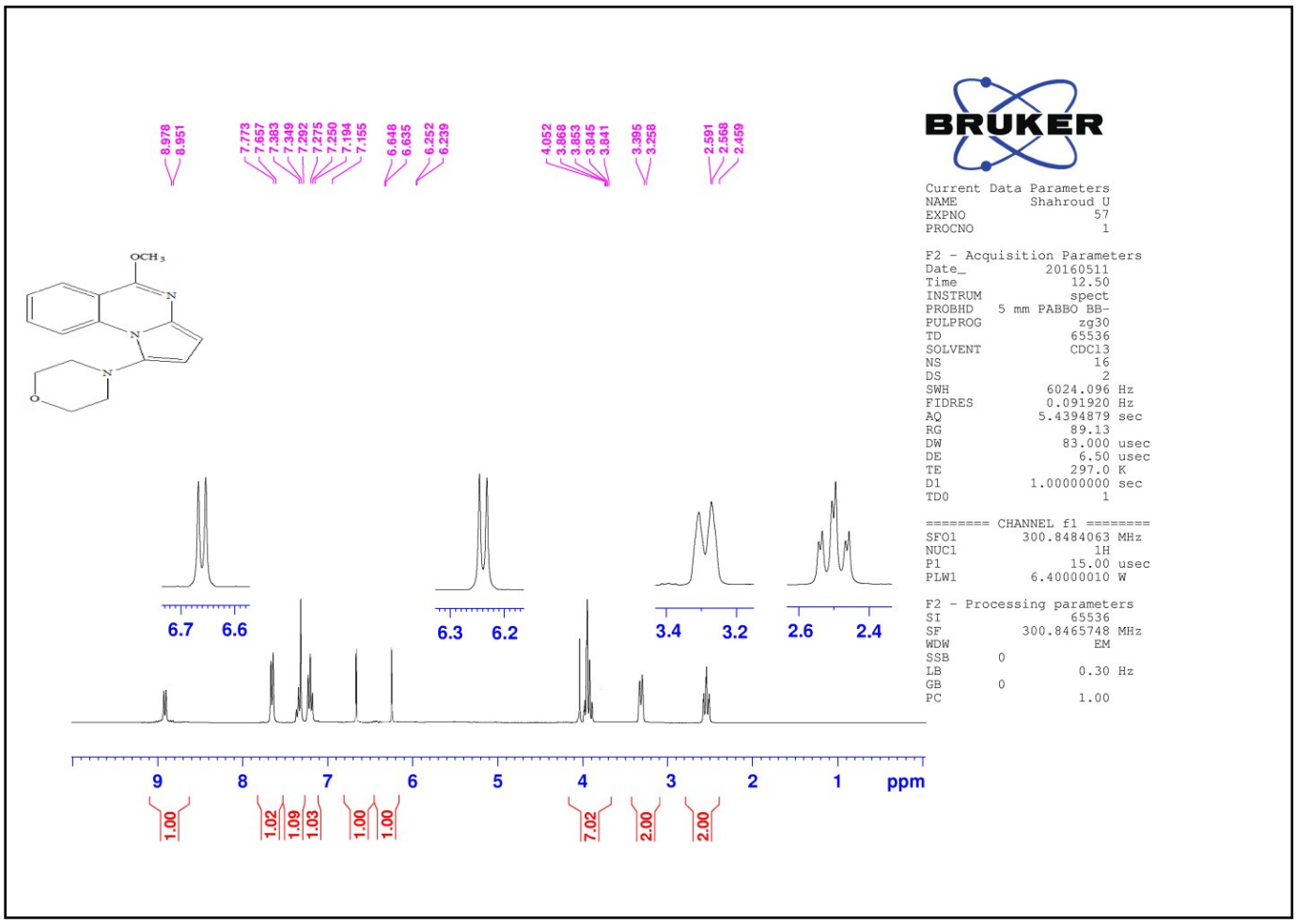

Figure 1. 300 MHz ^{1}H NMR spectrum of compound 4a in $CDCl_3$

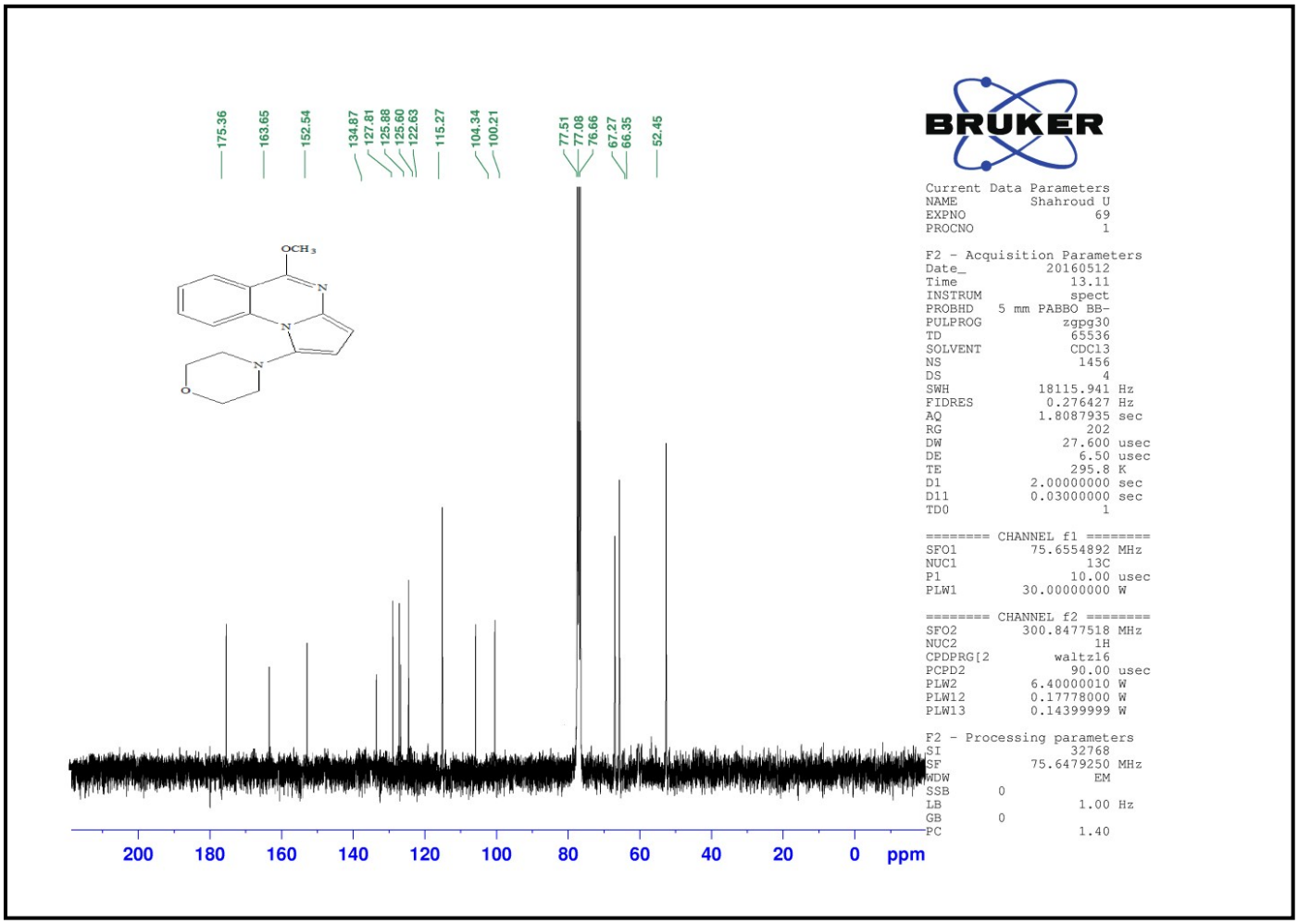

Figure 2. 75 MHz ^{13}C NMR spectrum of compound 4a in $CDCl_3$

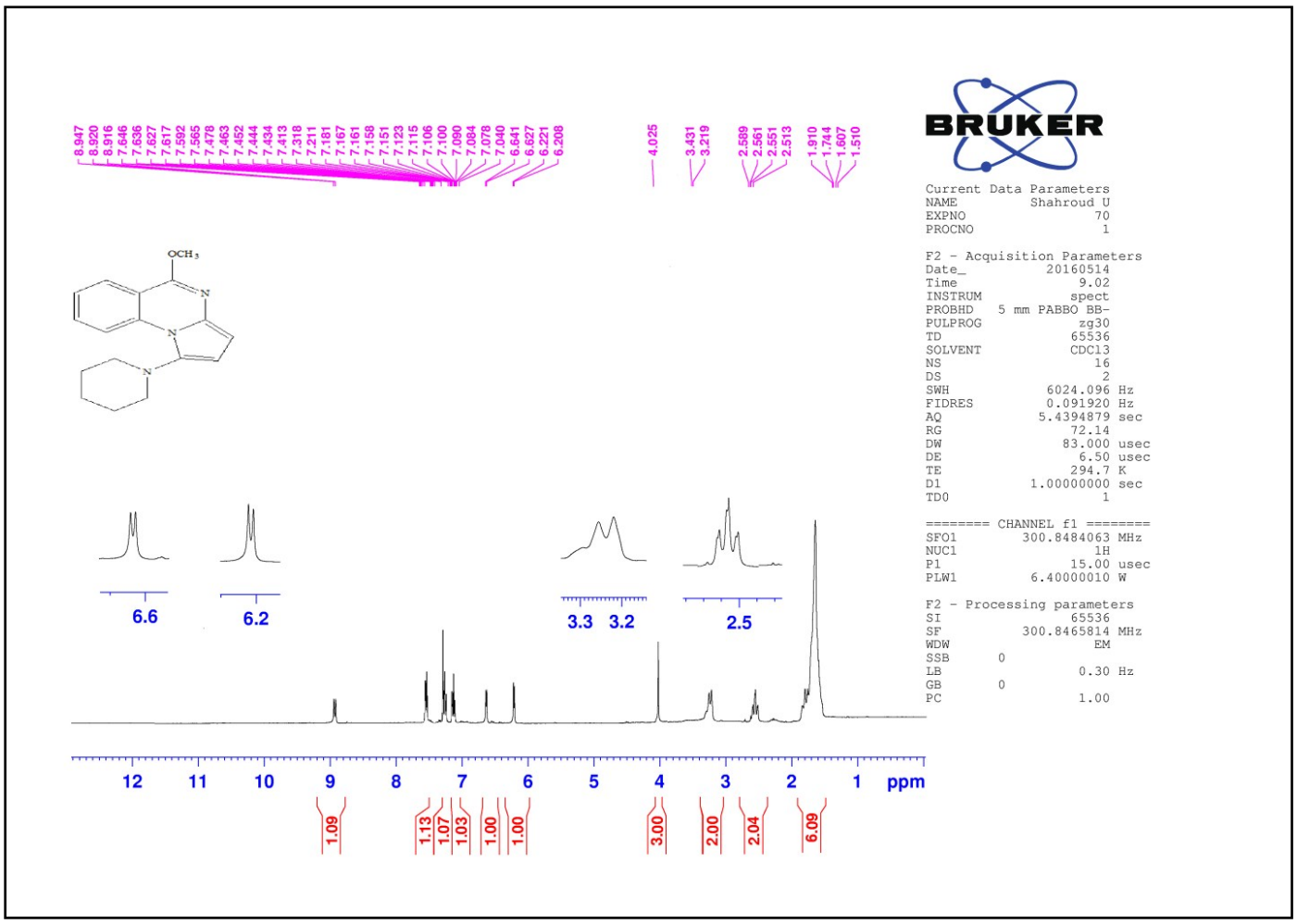

Figure 3. 300 MHz ^{1}H NMR spectrum of compound 4b in $CDCl_3$

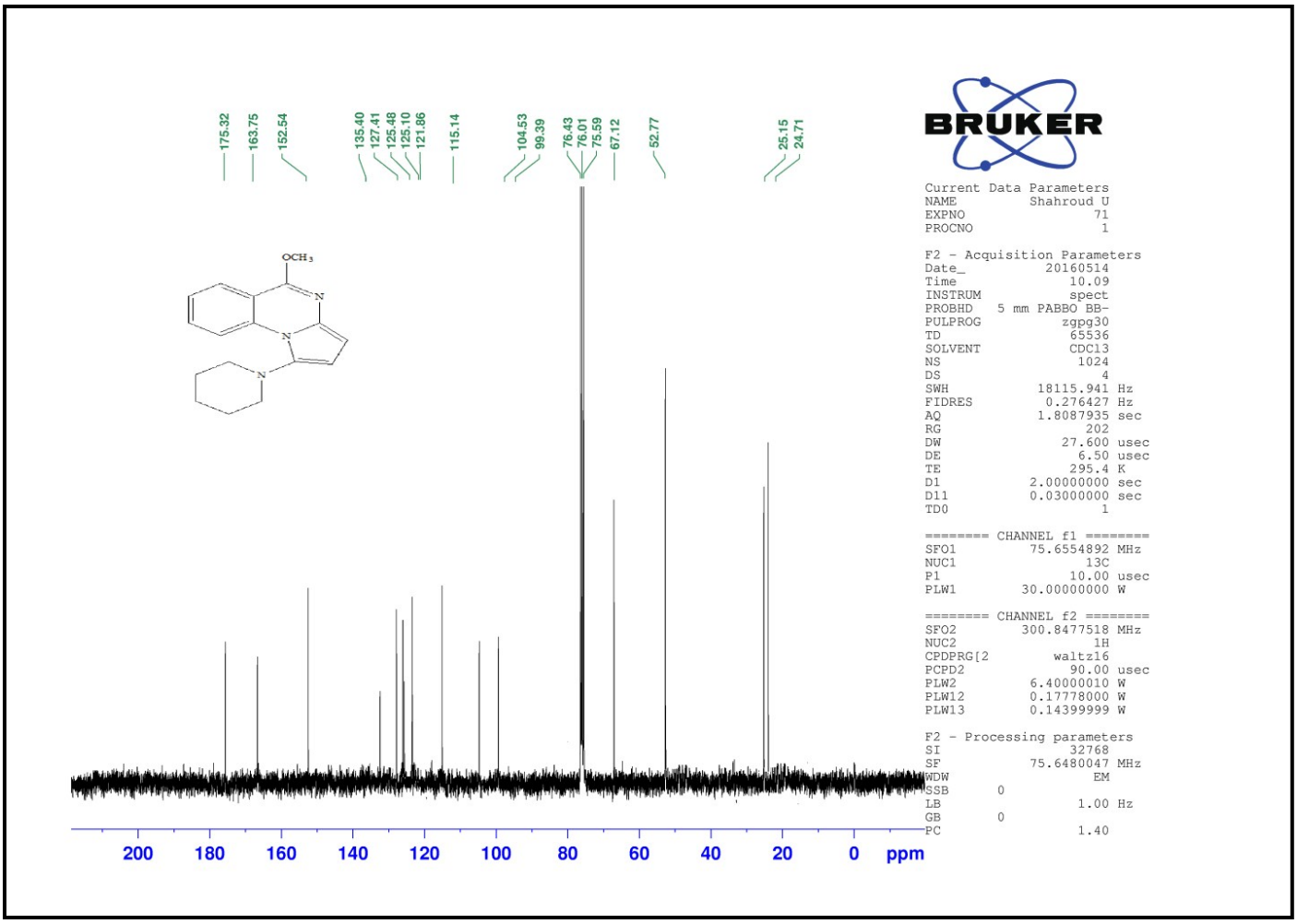

Figure 4. 75 MHz ^{13}C NMR spectrum of compound 4b in $CDCl_3$

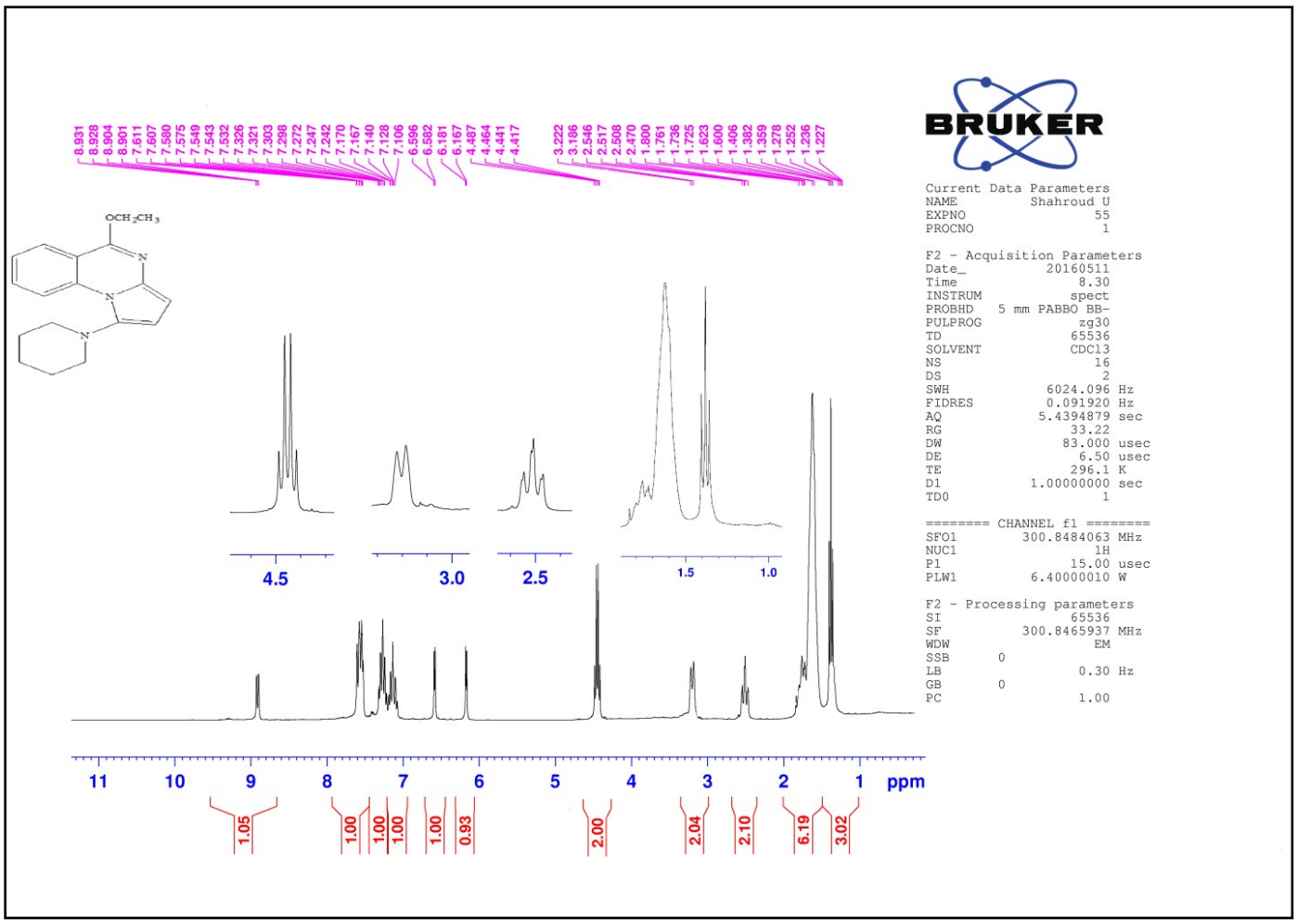

Figure 5. 300 MHz ^{1}H NMR spectrum of compound 4c in $CDCl_3$

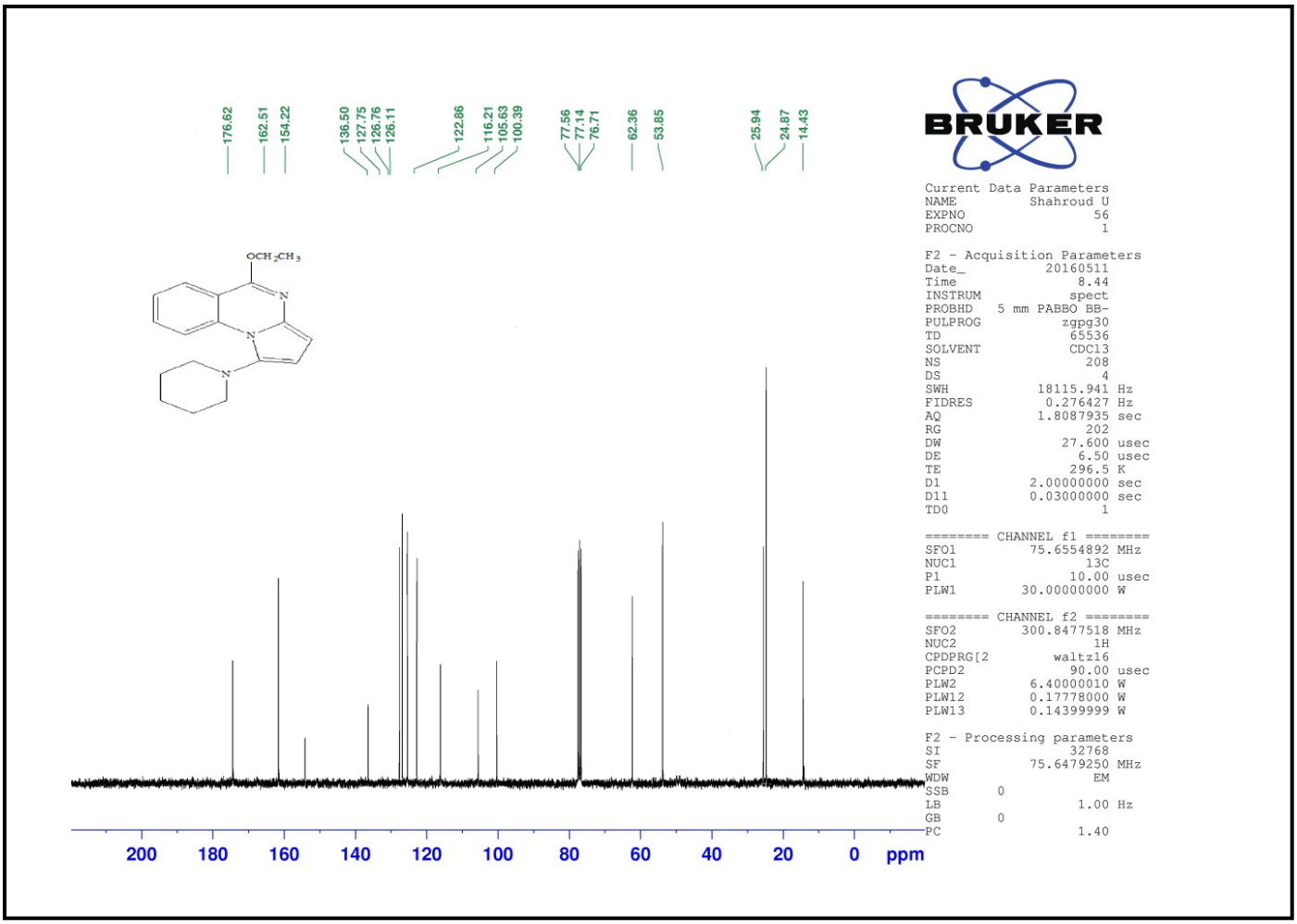

Figure 6. 75 MHz ^{13}C NMR spectrum of compound 4c in $CDCl_3$

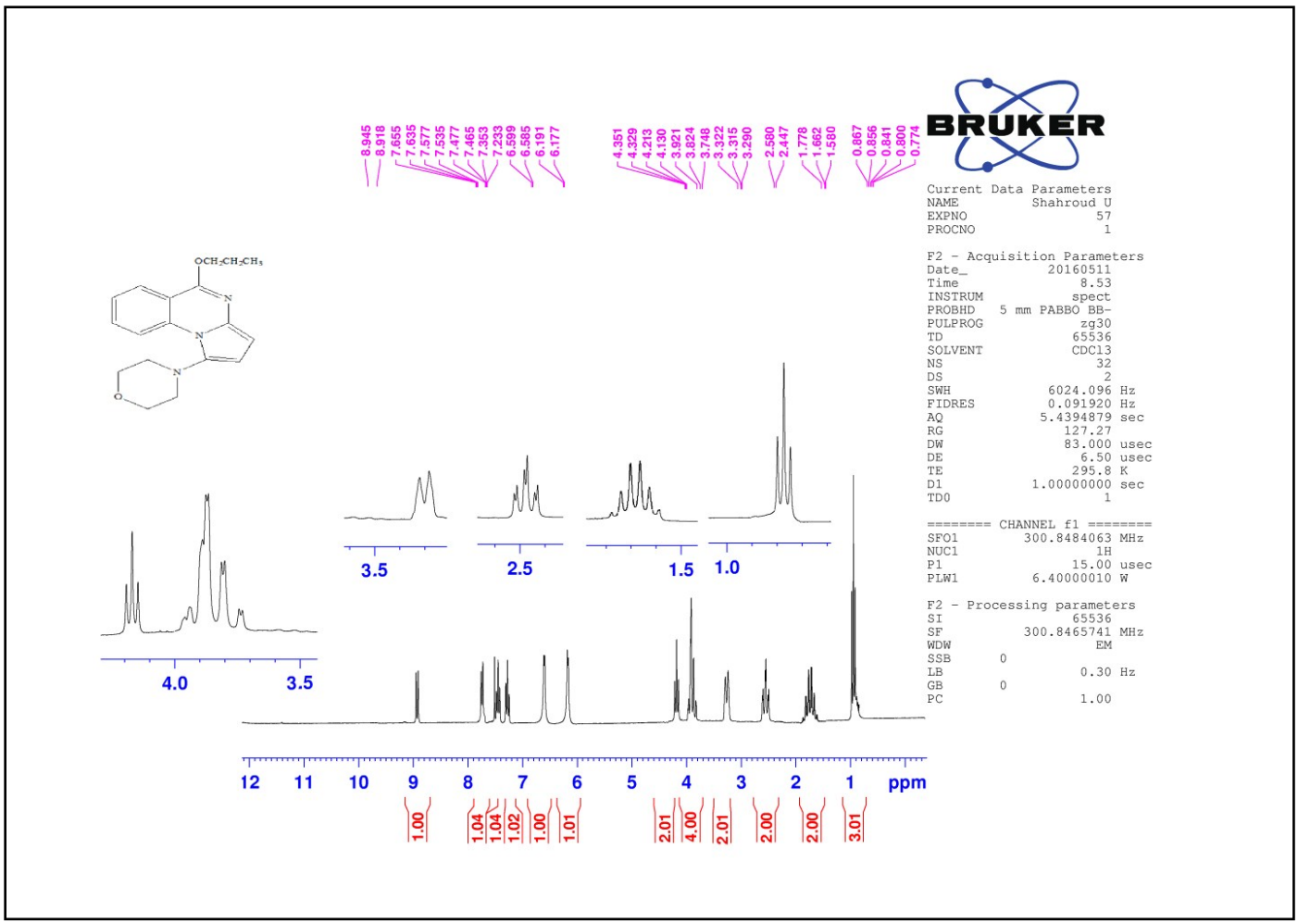

Figure 7. 300 MHz ^{1}H NMR spectrum of compound 4d in $CDCl_3$

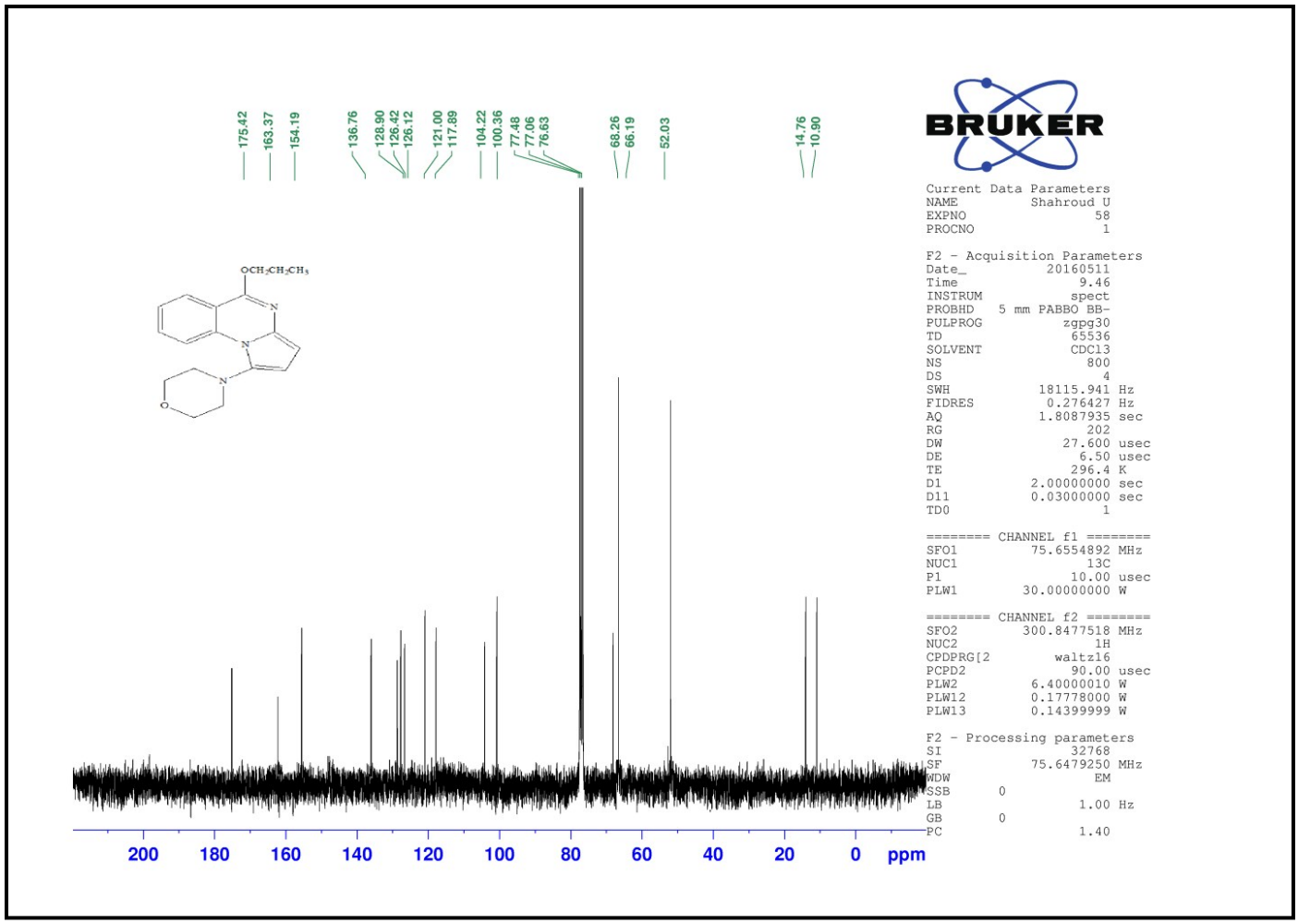

Figure 8. 75 MHz ^{13}C NMR spectrum of compound 4d in $CDCl_3$

Figure 9. 300 MHz ^{1}H NMR spectrum of compound 4e in $CDCl_3$

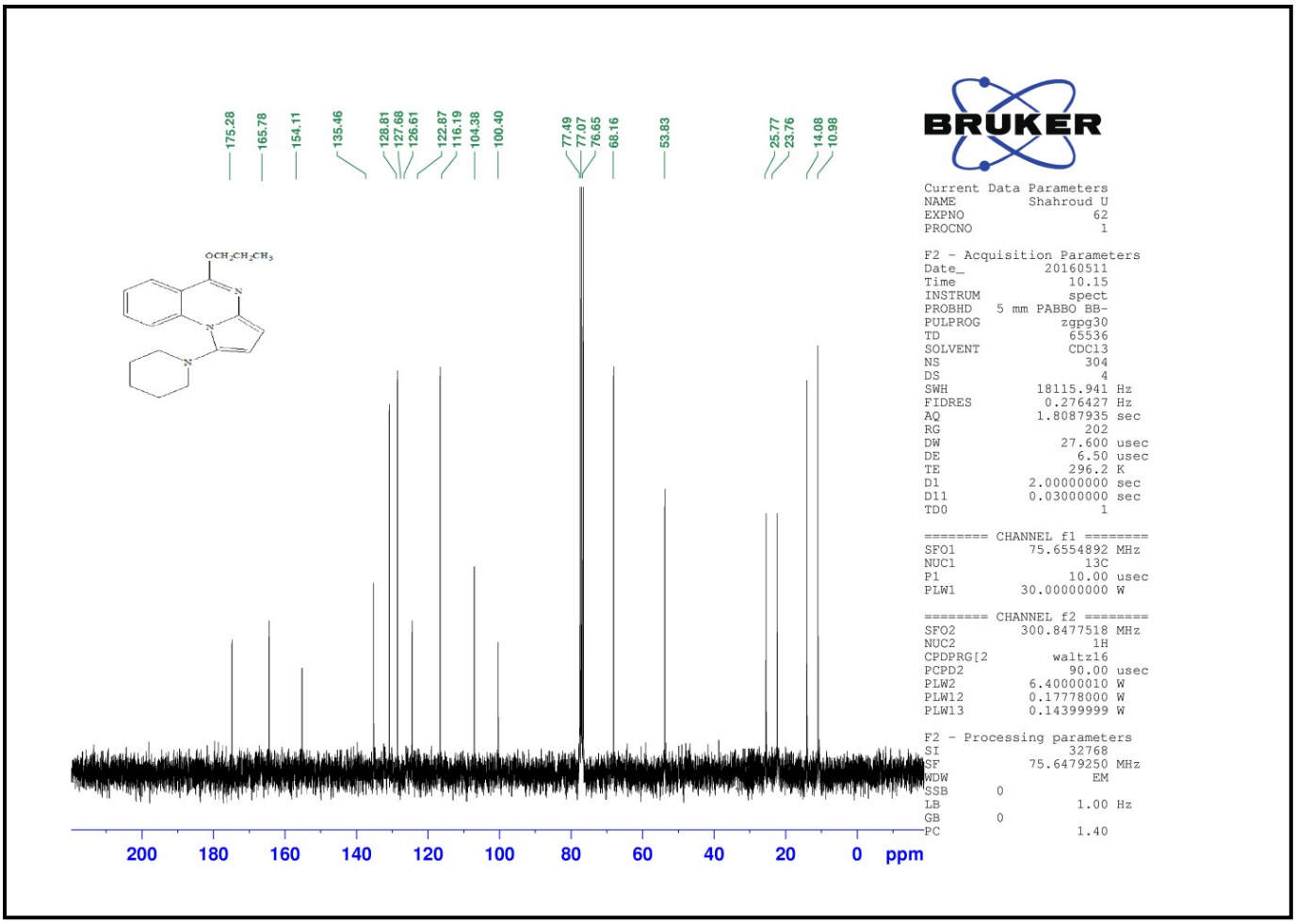

Figure 10. 75 MHz ^{13}C NMR spectrum of compound 4e in $CDCl_3$

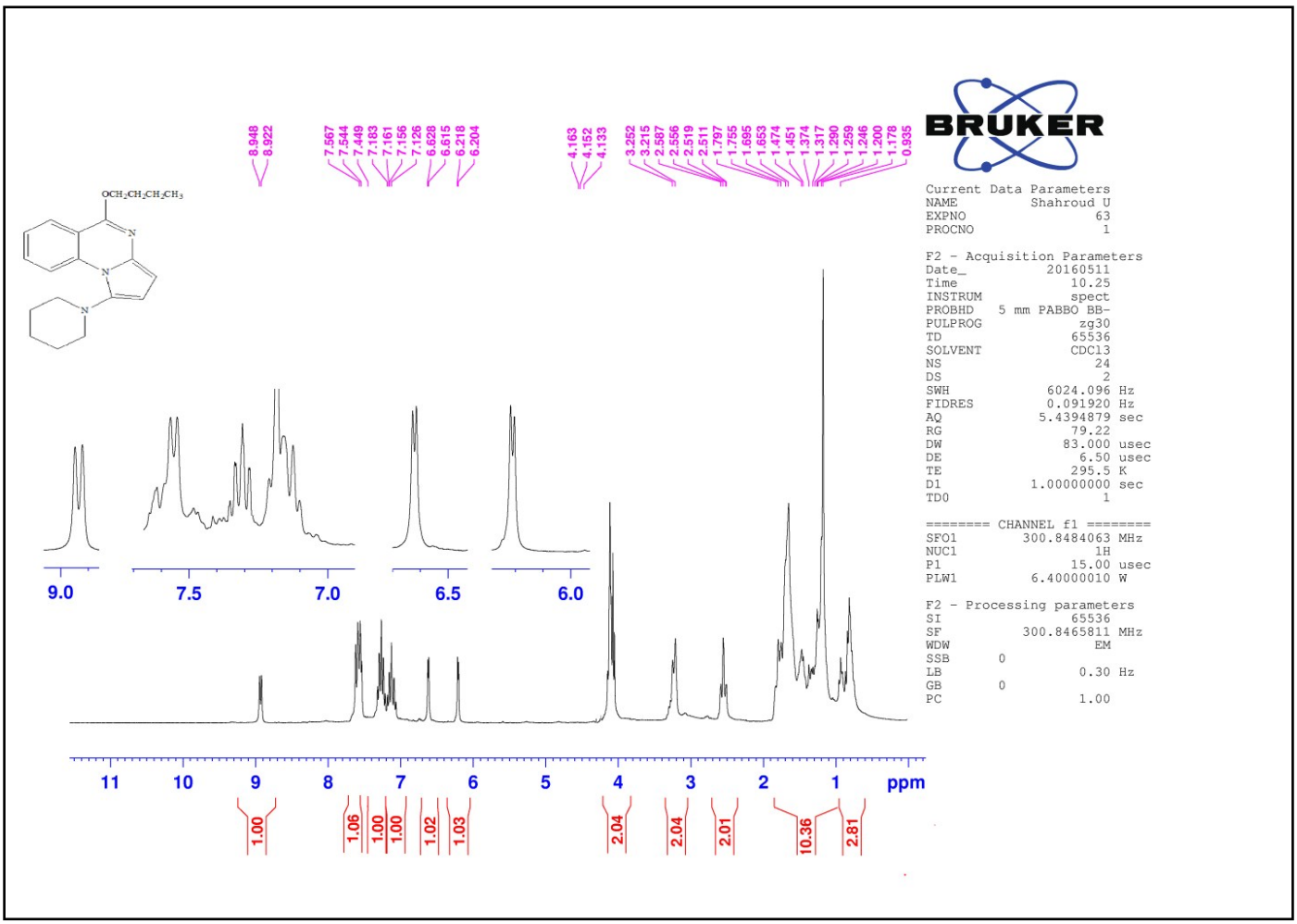

Figure 11. 300 MHz ^{1}H NMR spectrum of compound 4f in $CDCl_3$

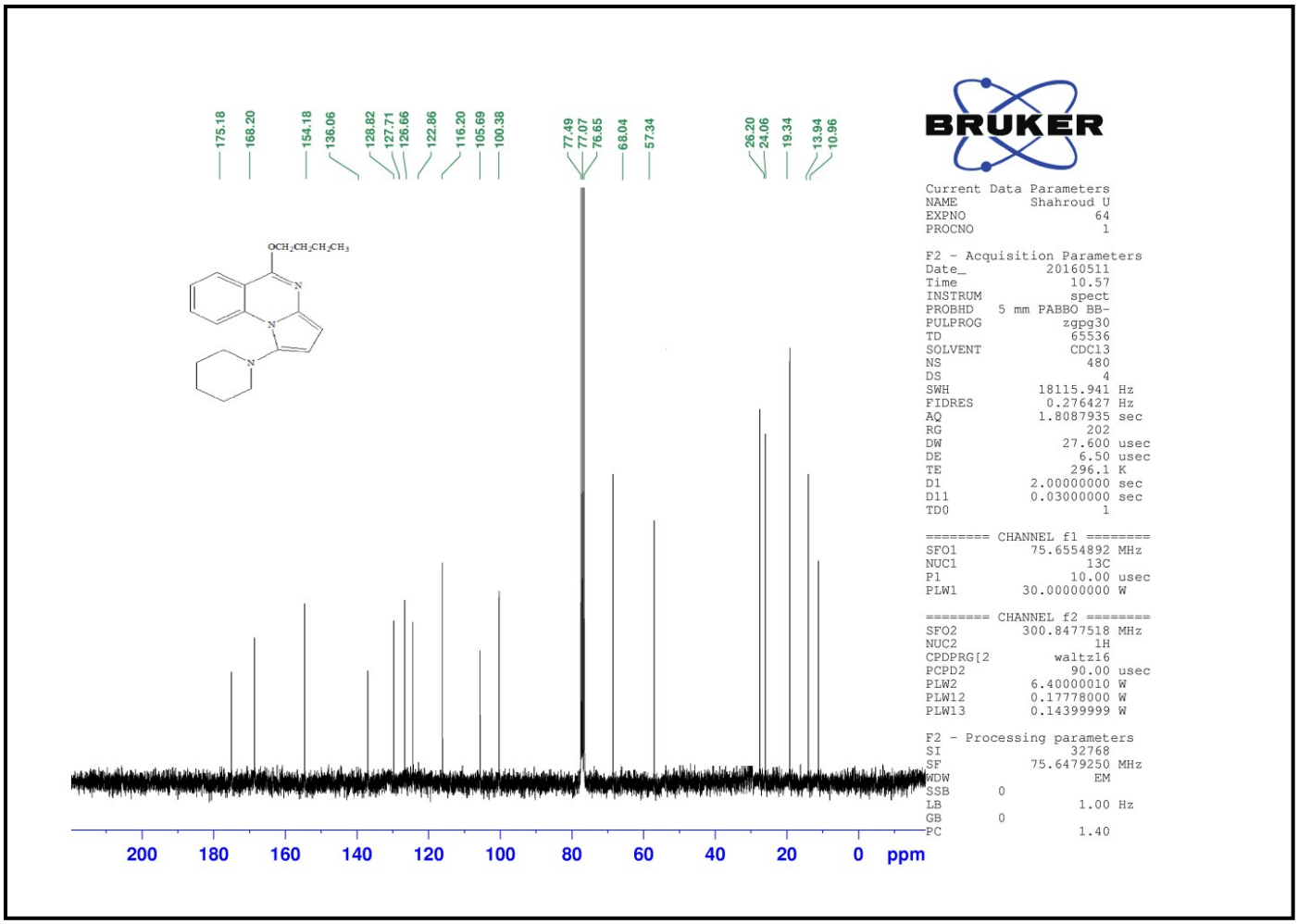

Figure 12. 75 MHz ^{13}C NMR spectrum of compound 4f in $CDCl_3$

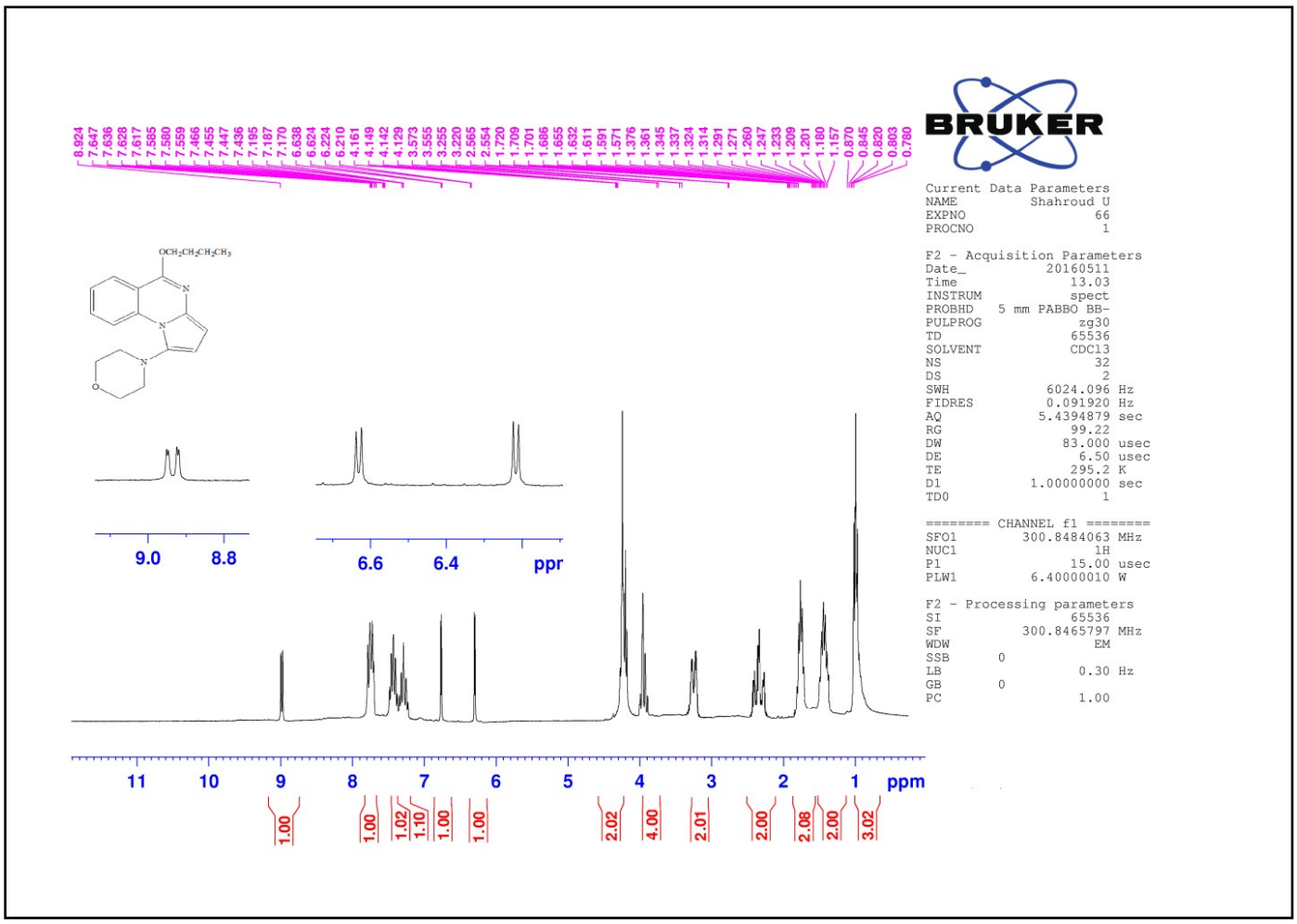

Figure 13. 300 MHz ^{1}H NMR spectrum of compound 4g in $CDCl_3$

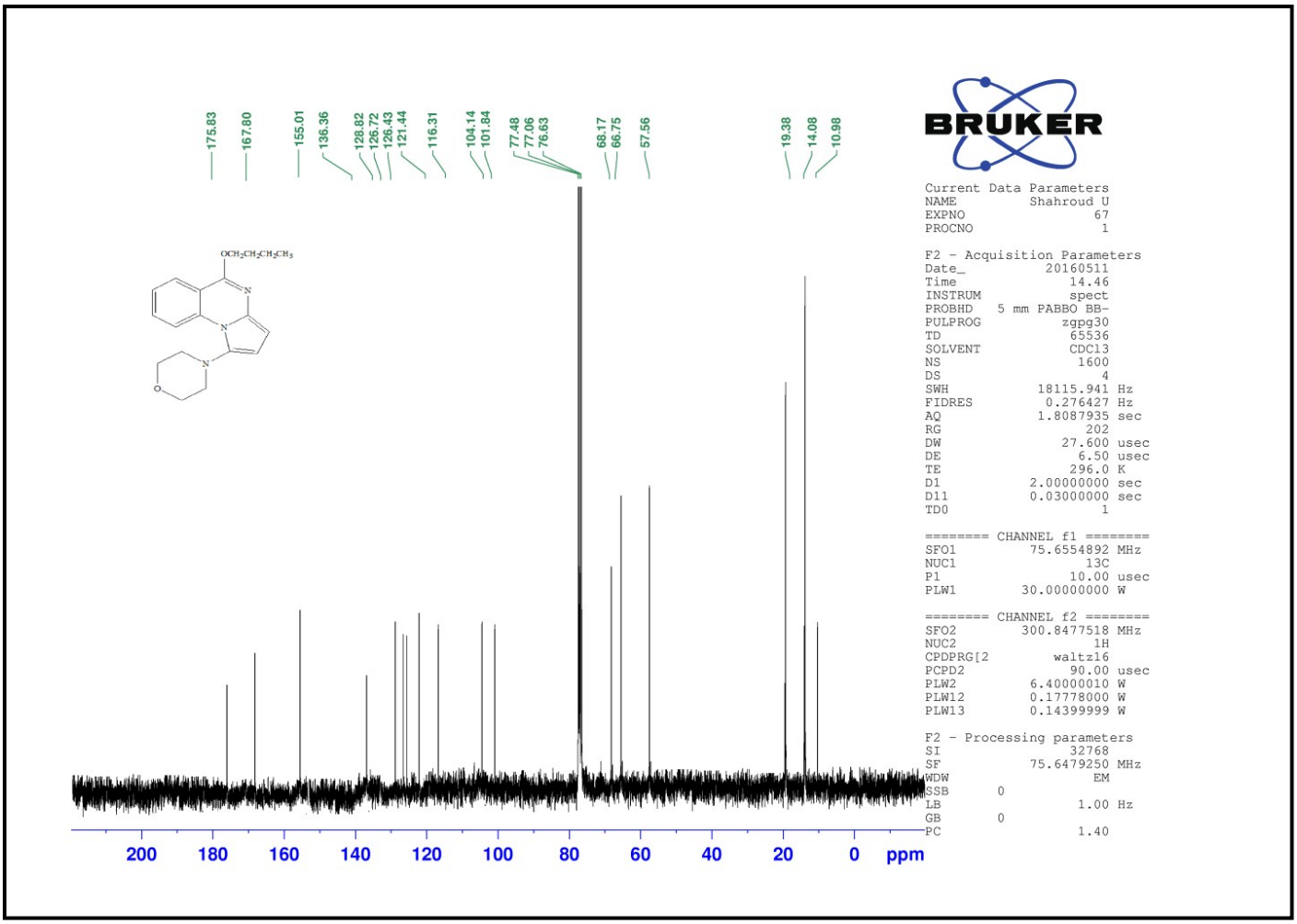

Figure 14. 75 MHz ^{13}C NMR spectrum of compound 4g in $CDCl_3$

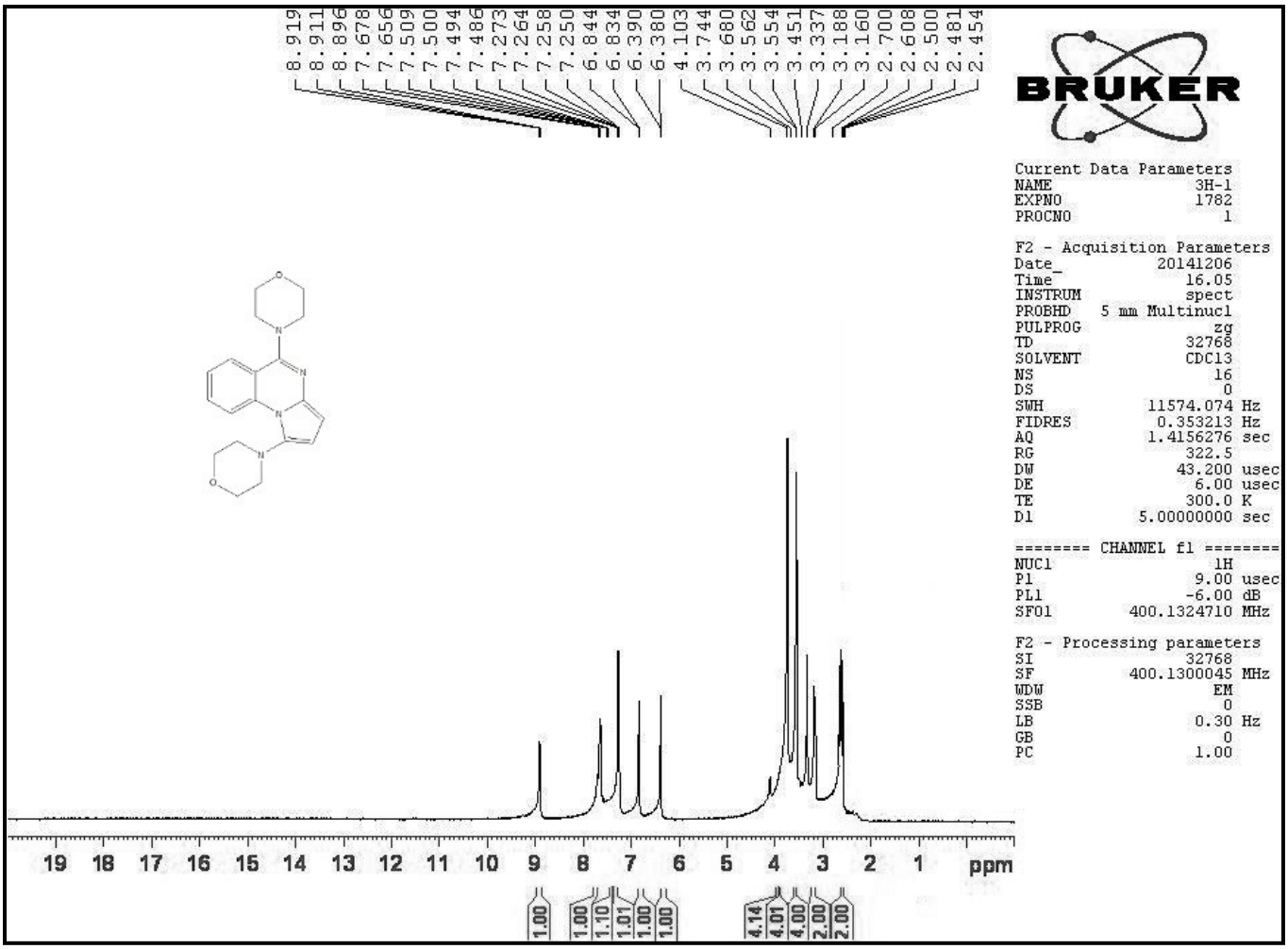

Figure 15. 300 MHz ^{1}H NMR spectrum of compound 4h in $CDCl_3$

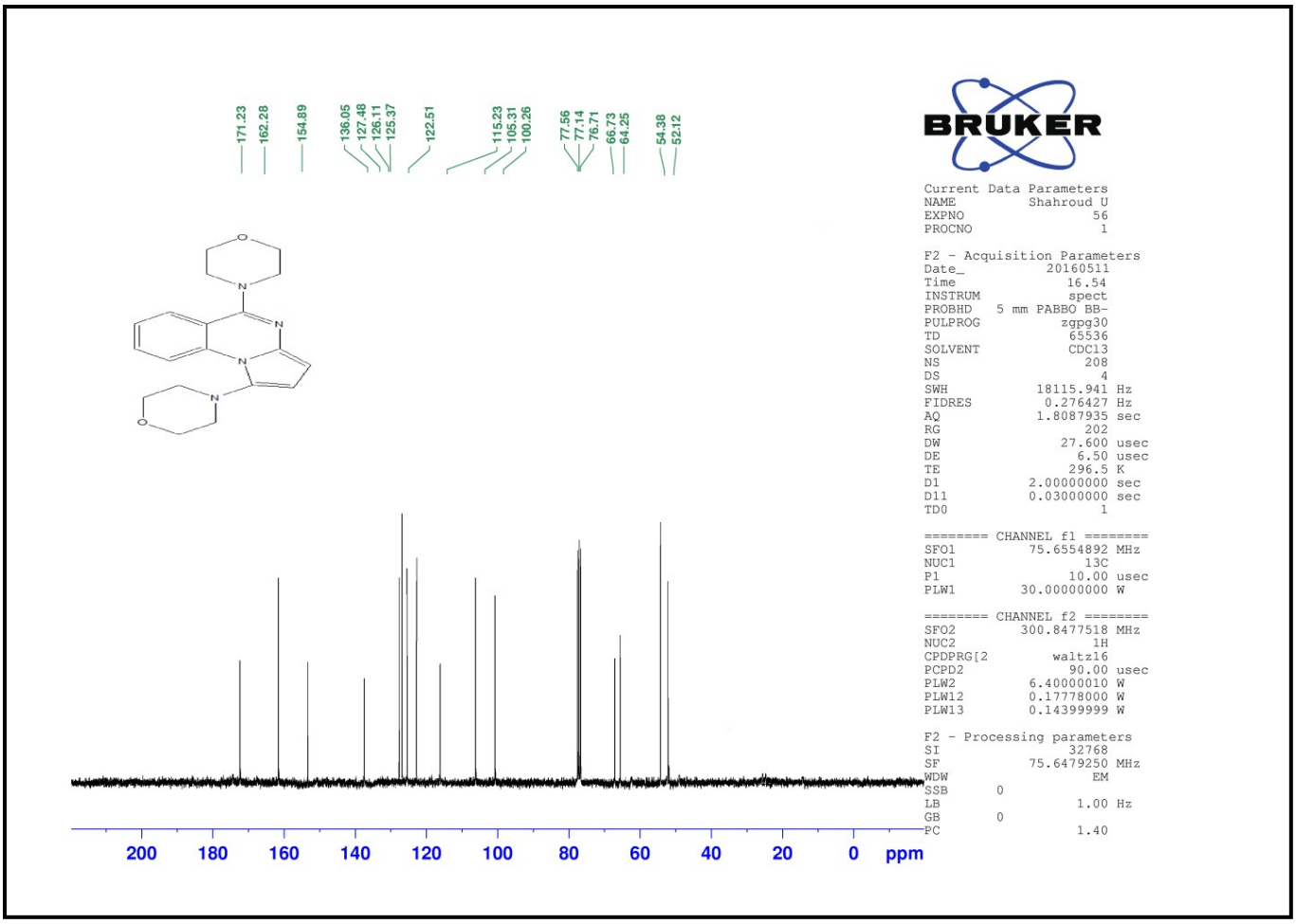

Figure 16. 75 MHz ^{13}C NMR spectrum of compound 4h in $CDCl_3$

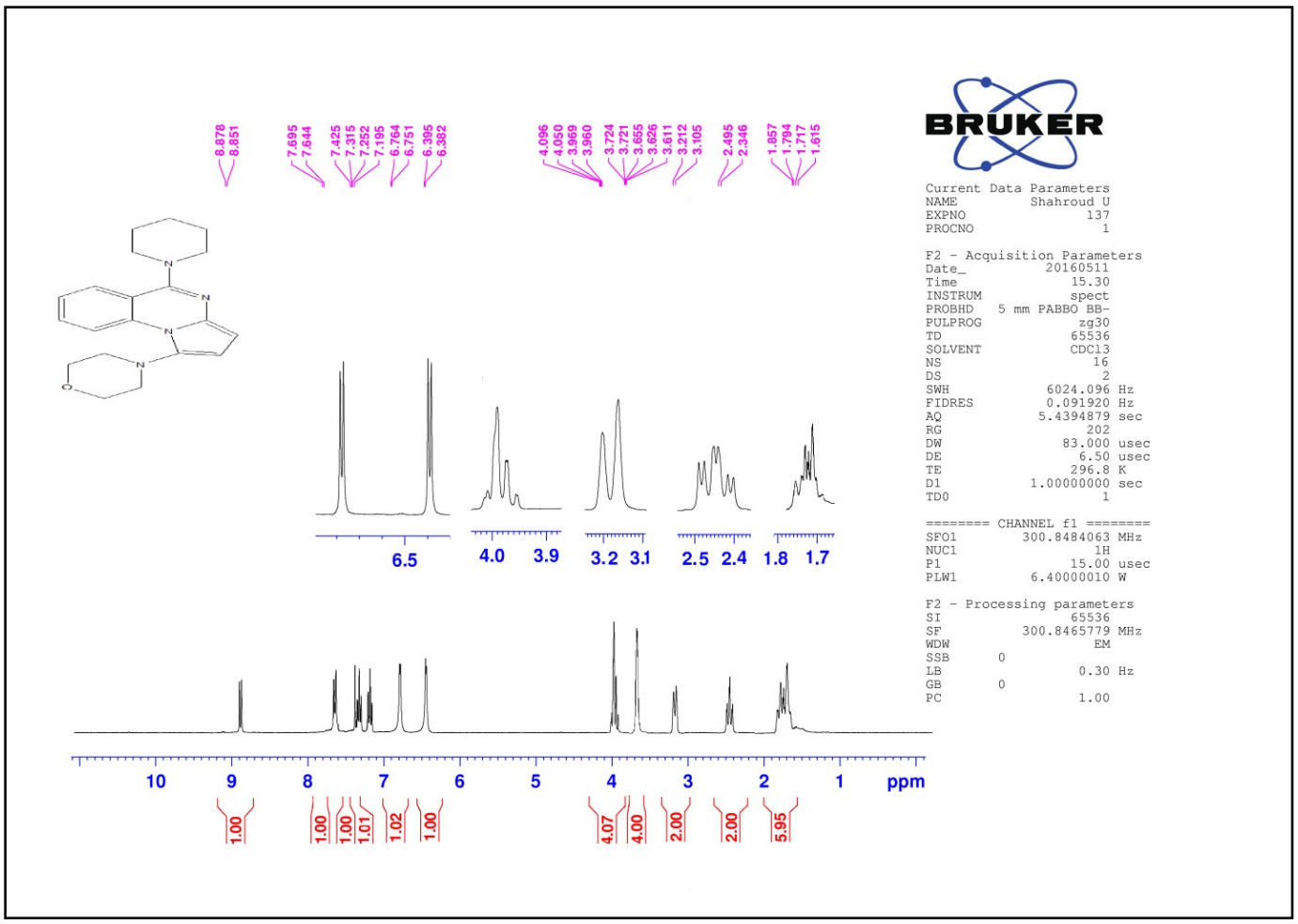

Figure 17. 300 MHz ^{1}H NMR spectrum of compound 4i in $CDCl_3$

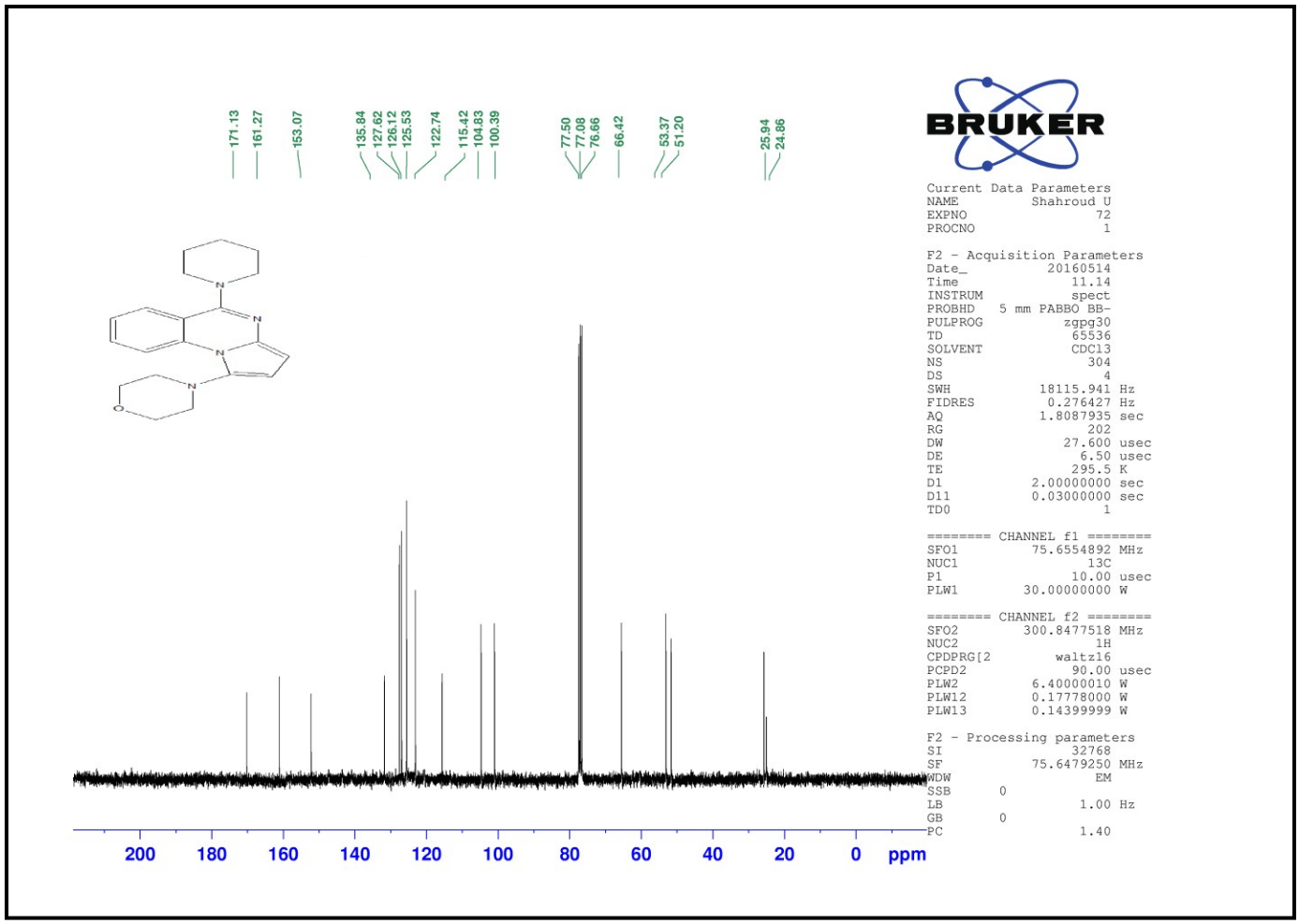

Figure 18. 75 MHz ^{13}C NMR spectrum of compound 4i in $CDCl_3$

References

[1]. Desai, PS.; Naik, PJ.; *Discovery*, **2014**, 17, 7.

[2]. Sagiv-Barfi, I.; Weiss, E.; Levitzki, A.; *Bioorganic & Medicinal Chemistry* , **2010**, 18, 6404.

[3]. Wang, H.; Wei, Ch.; Deng, X.; Li, F.; Quan, Zh.; *Arch. Pharm. Chem. Life Sci*. **2009**, 342, 671.