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Bis-heterocyclic systems based on 1,2,4-triazole-3-carboxamide as new anti-SARS-CoV2 agents

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General

All reactions were carried out using glassware with magnetic stirring. All chemicals were purchased from Merck (USA) and used without additional purification. All solvents were dried according to known methods[S1] and distilled before use, anhydrous methanol and ethanol were prepared by standard methods.[S1] ^{1}H and ^{13}C NMR spectra were recorded in $CDCl_3$ or DMSO-d_6 on a DPX-300 spectrometer (Bruker, Germany) at 25 °C. ^{1}H and ^{13}C NMR spectra were recorded at an operating frequency of 300 and 75 MHz respectively. Chemical shifts are presented in ppm relative to the solvent $CDCl_3$ (7.26 ppm), DMSO-d_6 (2.50 ppm) for ^{1}H NMR and $CDCl_3$ (77.0 ppm), DMSO-d_6 (39.5 ppm) for ^{13}C NMR. Coupling constants (*J*) are given in Hertz (Hz). The following abbreviations are used: s = singlet, d = doublet, t = triplet, m = multiplet, q = quartet, and br.s = broad singlet. High-resolution mass spectra (HRMS) were recorded on Agilent 6224 (USA) using electron spray ionization (ESI). The specific rotation $[\alpha]^{27}_{D}$ was measured on an Atago AP-300 automatic polarimeter (Russia). Solvents were removed on a Heidolph LABOROTA 4000 rotary vacuum evaporator (Germany) with a water jet pump. Melting points were determined on the Cole-Parmer MP-200D-120 device in an open capillary.

Sorbfil silica gel plates (Russia) were used for thin-layer chromatography. Substances visualization on thin-layer chromatograms were carried out using UV irradiation at 254 nm and iodine followed by phosphomolybdic acid or ninhydrin. Column chromatography was carried out on silica gel Kiselgel 60 0.040-0.063 mm (Merck, Germany).

LCMS measurements were performed in the Agilent InfinityLab LC/MSD iQ chromatographic system (USA) consisting of: Agilent 1260 Infinity II SFC Control Module, Agilent 1260 Infinity High Performance Degasser, Agilent 1260 Infinity II SFC Quaternary Pump, Agilent 1260 Infinity II Multisampler, Agilent 1260 Infinity II Multicolumn Thermostat , and Agilent Single Quadrupole LC/MSD iQ mass Spectrometric detector (m/z range 2-1450). Separation was performed on an Agilent Poroshell 300SB-C18 chromatographic column, 2.1 x 75 mm, 5 um.

Separation conditions:

A: 0.1% formic acid in MeCN;

B: 0.1% formic acid in H_2O.

The elution mode was gradient:

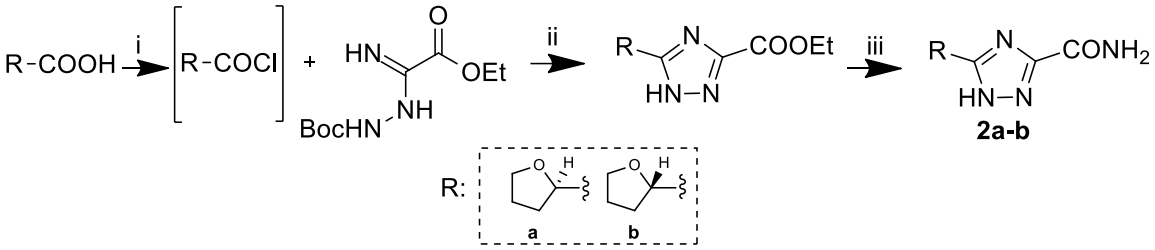
0 min – 10% A and 90% B;

10 min – 95% A and 5% B;

The volume of the injected sample – 5 μl. The flow rate of the mobile phase – 0.5 ml/min. The column temperature – 30 °C. ESI ionization ±3.5 kV, spray gas – nitrogen.

Synthetic procedures

Enantiomers of 5-tetrahydrofuran-2yl 1,2,4-triazole-3-carboxamides synthesis



Scheme S1 *Reagents and conditions:* i, $SOCl_2$, r.t.; ii, Py (3eq), toluene, reflux; iii, NH_3, MeOH, reflux.

General synthetic procedure for enantiomers of ethyl 5-(tetrahydrofuran-2-yl)-1,2,4-triazole-3-carboxylates

To a suspension of optically pure acid (purchased from Sigma Aldrich) 2 eq. of thionyl chloride was added drop by drop on an ice bath. After the addition was complete, the reaction mixture was stirred at room temperature for 2.5 hours. The conversion of the acid was monitored by TLC (10% methanol in chloroform). After complete conversion of the acid, the volatile components were evaporated. The resulting oil was used as corresponding carboxylic acid chloride without further purification.

Anhydrous pyridine (3 equiv., in this procedure hereinafter eq. are given to oxalamidrazone) was added to a suspension of ethyl β-*N*-Boc-oxalamidrazone in anhydrous toluene followed by a dropwise addition of 2.15 eq. corresponding carboxylic acid chloride. The reaction mixture was stirred at 90°C for 12 h. After complete conversion of ethyl β-*N*-Boc-oxalamidrazone (controled by TLC, eluent 2% methanol in chloroform), the reaction was concentrated on a rotary evaporator. Then H_2O (5 ml) was added, and the resulting mixture was extracted with EtOAc (3×10 ml). The combined organic phases were dried over anhydrous Na_2SO_4, filtered off and the solvent was evaporated. The product was isolated by column chromatography on silica gel, eluent toluene–acetone, gradient of acetone from 0 to 15%.

Ethyl 5-((R)-tetrahydrofuran-2-yl)-1,2,4-triazole-3-carboxylate was obtained by the general procedure from (*R*)-tetrahydrofuran-2-carboxylic acid (2.55 g, 22 mmol), thionyl chloride (3.19 ml, 44 mmol), anhydrous pyridine (2.41 ml), anhydrous toluene (15 ml), and ethyl β-N-Boc-oxalamidrazone (2.37 g, 10 mmol). Yield 0.43 g (20%), product was obtained as white crystals.

R_f = 0.65 (ethyl acetate). M.p. 74-75°C. ^{1}H NMR ($CDCl_3$): 1.35 (3H, t, J = 7.1, $CH_2C\underline{H_3}$); 1.89–2.02 (2H, m, $C\underline{H_2}C\underline{H_2}CH$); 2.20–2.29 (1H, m, $OCHC\underline{H_2}$); 2.35–2.47 (1H, m, $C\underline{H_2}CH_2O$); 3.87–4.03 (2H, m, $C\underline{H_2}O$); 4.41 (2H, q, J = 7.0, $C\underline{H_2}CH_3$); 5.13–5.20 (1H, m, $OC\underline{H}CH_2$). ^{13}C NMR ($CDCl_3$): 14.1; 25.5; 31.7; 61.9; 69.3; 73.7; 154.4; 159.7; 160.3. HRMS: for $C_9H_{13}N_3O_3$ $[M+H]^+$ calculated: 212.1035, found: 212.1039. $[\alpha]^{27}_D$ +29±0.2°.

Ethyl 5-((S)-tetrahydrofuran-2-yl)-1,2,4-triazole-3-carboxylate was obtained by the general procedure from (*S*)-tetrahydrofuran-2-carboxylic acid (4.99 g, 43 mmol), thionyl chloride (10.23 ml, 86 mmol), anhydrous pyridine (4.84 ml), anhydrous toluene (15 ml), and ethyl β-N-Boc-oxalamidrazone (4.63 g, 20 mmol). Yield 1.14 g (27%), product was obtained as white crystals.

R_f = 0.65 (ethyl acetate). M.p. 76-77°C. ^{1}H NMR ($CDCl_3$): 1.35 (3H, t, J = 7.1, $CH_2C\underline{H_3}$); 1.86–2.06 (2H, m, $C\underline{H_2}C\underline{H_2}CH$); 2.18–2.28 (1H, m, $OCHC\underline{H_2}$); 2.35–2.47 (1H, m, $C\underline{H_2}CH_2O$); 3.87–4.02 (2H, m, $C\underline{H_2}O$); 4.41 (2H, q, J = 7.0, $C\underline{H_2}CH_3$); 5.13–5.19 (1H, m, $OC\underline{H}CH_2$). ^{13}C NMR ($CDCl_3$): 14.1; 25.5; 31.7; 61.9; 69.3; 73.7; 154.4; 159.7; 160.3. HRMS: for $C_9H_{13}N_3O_3$ $[M+H]^+$ calculated: 212.1035, found: 212.1037. $[\alpha]^{27}_D$ -29±0.2°.

General synthetic procedure for enantiomers of 5-(tetrahydrofuran-2-yl)-1,2,4-triazole-3-carboxamides (2)

Ethyl 5-(tetrahydrofuran-2-yl)-1,2,4-triazole-3-carboxylate was dissolved in 1.5 ml of 10M NH_3 solution in anhydrous methanol. The mixture was heated under reflux, and 0.5 ml of 10M NH_3 solution in anhydrous methanol was added every 6 h. Upon completion conversion of the ester (TLC control, eluent 5% methanol in chloroform), the solvent was removed on a rotary evaporator. The product was suspended in anhydrous Me_2CO, filtered, and dried in a desiccator under reduced pressure over NaOH for 12 h.

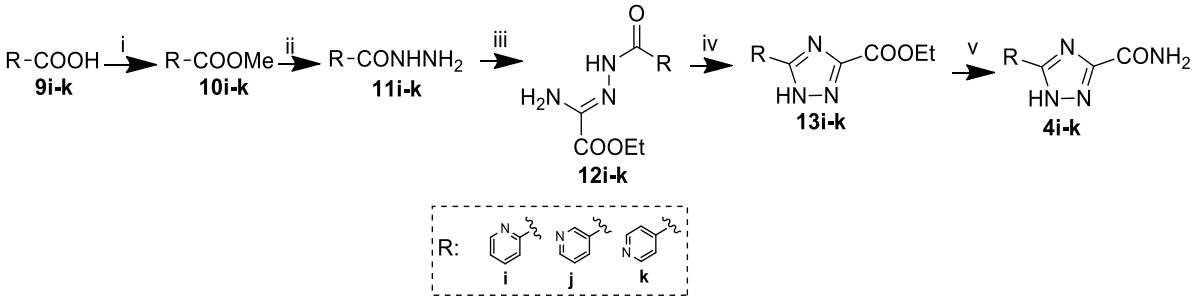
5-((R)-Tetrahydrofuran-2-yl)-1,2,4-triazole-3-carboxamide (2a) was obtained from ethyl 5-((*R*)-tetrahydrofuran-2-yl)-1,2,4-triazole-3-carboxylate (150 mg, 0.71 mmol) in 24 h. Yield 125 mg (97%), product was obtained as white crystals.

R_f = 0.45 (ethyl acetate). M.p. 128–130°C. ^{1}H NMR (DMSO-d_6): 1.89-2.00 (2H, m, OCHC$\underline{H_2}$C$\underline{H_2}$); 2.02-2.14 (1H, m, OCHCH$_2$C$\underline{H_2}$); 2.18-2.29 (1H, m, OCHC$\underline{H_2}$); 3.74-3.93 (2H, m, C$\underline{H_2}$O); 4.95-4.99 (1H, m, OC$\underline{H}$); 7.59 (1H, br. s, CON$\underline{H_2}$); 7.83 (1H, br. s, CON$\underline{H_2}$). ^{13}C NMR (DMSO-d_6): 25.3; 30.9; 68.0; 72.9; 154.0; 159.9; 160.6. HRMS: for $C_7H_{10}N_4O_2$ $[M+H]^+$ calculated: 183.0882, found: 183.0888. $[\alpha]^{27}_D$ +19±0.2°.

5-((S)-Tetrahydrofuran-2-yl)-1,2,4-triazole-3-carboxamide (2b) was obtained from 5-((*S*)-tetrahydrofuran-2-yl)-1,2,4-triazole-3-carboxylate (150 mg, 0.71 mmol) in 24 h. Yield 110 mg (85%), product was obtained as white crystals.

R_f = 0.45 (ethyl acetate). M.p. 130–132°C. ^{1}H NMR (DMSO-d_6): 1.86-1.99 (2H, m, OCHC$\underline{H_2}$C$\underline{H_2}$); 2.01-2.11 (1H, m, OCHCH$_2$C$\underline{H_2}$); 2.20-2.25 (1H, m, OCHC$\underline{H_2}$); 3.74-3.93 (2H, m, C$\underline{H_2}$O); 4.95-5.00 (1H, m, OC$\underline{H}$); 7.64 (1H, br. s, CON$\underline{H_2}$); 7.89 (1H, br. s, CON$\underline{H_2}$). ^{13}C NMR (DMSO-d_6 and CF_3COOH): 25.3; 31.0; 68.1; 72.9; 154.0; 159.9; 160.7. HRMS: for $C_7H_{10}N_4O_2$ $[M+H]^+$ calculated: 183.0882, found: 183.0889. $[\alpha]^{27}_D$ -19±0.2°.

5-Pyridyl-1,2,4-triazole-3-carboxamides 4i-k synthesis



Scheme S2 *Reagents and conditions:* i, $SOCl_2$, MeOH (anhydrous), r.t.; ii, N_2H_4*H_2O, MeOH, at r.t.; iii, NH_2SC-COOEt, EtOH, at r.t.; iv, o-xylene, reflux; v, NH_3, MeOH, reflux.

General synthetic procedure for methyl pyridine carboxylates (10)

To a suspension of pyridine carboxylic acid **9** in anhydrous methanol (20 ml per 2 g), 3 equiv. of thionyl chloride was added drop by drop on an ice bath. After the addition was complete, the reaction mixture was stirred at room temperature for 4 hours. The volatile components were evaporated. The residue was dissolved in 20 ml of ethyl acetate and neutralized with a saturated aqueous solution of Na_2CO_3. The organic layer was dried with anhydrous Na_2SO_4, filtered off and evaporated.

Methyl pyridine-2-carboxylates (10i) was obtained from 4.0 g (32.5 mmol) of pyridine-2-carboxylic acid **9i**, 7.1 ml (97.5 mmol) of thionyl chloride, 4.4 g (99%) of product **10i** was obtained as yellow oil.

R_f = 0.68 (5% methanol in chloroform). ^{1}H NMR ($CHCl_3$) δ: 4.00 (3H, s, -O-CH_3); 7.45-7.50 (1H, m, pyridine ring 5-CH); 7.87-7.81 (1H, m, pyridine ring 4-CH); 8.11-8.14 (1H, m, pyridine ring 3-CH); 8.73-8.75 (1H, m, pyridine ring 6-CH). ^{13}C NMR ($CHCl_3$) δ: 52.9; 125.2; 127.0; 137.4; 147.6; 149.5; 165.3.

Methyl pyridine-3-carboxylates (10j) was obtained from 4.0 g (32.5 mmol) of pyridine-3-carboxylic acid **9j**, 7.1 ml (97.5 mmol) of thionyl chloride, 4.4 g (99%) of product **10j** was obtained as yellow oil.

R_f = 0.65 (5% methanol in chloroform). ^{1}H NMR ($CHCl_3$) δ: 3.92 (3H, s, -O-CH_3); 7.35-7.39 (1H, m, pyridine ring 5-CH); 8.25-8.30 (1H, m, pyridine ring 4-CH); 8.73-8.75 (1H, m, pyridine ring 6-CH); 9.19 (1H, m, pyridine ring 2-CH). ^{13}C NMR ($CHCl_3$) δ: 52.3; 123.3; 126.1; 137.1; 150.6; 153.1; 165.5.

Methyl pyridine-4-carboxylates (10k) was obtained from 4.0 g (32.5 mmol) of pyridine-4-carboxylic acid **9k**, 7.1 ml (97.5 mmol) of thionyl chloride, 4.4 g (99%) of product **10k** was obtained as yellow oil.

R_f = 0.70 (5% methanol in chloroform). ^{1}H NMR ($CHCl_3$) δ: 3.95 (3H, s, -O-CH_3); 7.85-7.87 (2H, m, pyridine ring 3-CH and 5-CH); 8.76-8.78 (1H, m, pyridine ring 2-CH and 6-CH). ^{13}C NMR ($CHCl_3$) δ: 52.7; 123.0; 137.8; 150.0; 165.3.

General synthetic procedure for pyridine carbohydrazides (11)

Hydrazine hydrate (3 eq.) was added to a solution methyl pyridinecarboxylate **10** in 10 ml methanol. The reaction mass was stirred at room temperature for 6 hours. Then the volatile components were evaporated. The residue was suspended in 15 ml of toluene and re-evaporated twice.

Pyridine-2-carboxylic acid hydrazide (11i) was obtained from 4.4 g (32.1 mmol) **10i**, 4.7 ml (96.3 mmol) of hydrazine hydrate, 4.1 g (94%) of product **11i** was obtained as white crystals.

R_f = 0.35 (10% methanol in chloroform). Mp 110°C. ^{1}H NMR (DMSO-d_6) δ: 4.55 (2H, s, -NH-N$\underline{H}_2$); 7.53-7.57 (1H, m, pyridine ring 5-CH); 7.93-7.97 (2H, m, pyridine ring 3-CH and 4-CH); 8.58-8.60 (1H, m, pyridine ring 6-CH); 9.83 (1H, s, -N$\underline{H}$-NH_2). ^{13}C NMR (DMSO-d_6) δ: 121.6; 126.1; 137.5; 148.4; 149.8; 162.6.

Pyridine-3-carboxylic acid hydrazide (11j) was obtained from 4.4 g (32.1 mmol) **10j**, 4.7 ml (96.3 mmol) of hydrazine hydrate, 3.5 g (80%) of product **11j** was obtained as white crystals.

R_f = 0.38 (10% methanol in chloroform). Mp 161°C. ^{1}H NMR (DMSO-d_6) δ: 4.55 (2H, s, -NH-N$\underline{H}_2$); 7.46-7.50 (1H, m, pyridine ring 5-CH); 8.13-8.15 (1H, m, pyridine ring 4-CH); 8.67-8.69 (1H, m, pyridine ring 6-CH); 8.95-8.96 (1H, m, pyridine ring 2-CH); 9.95 (1H, s, -N$\underline{H}$-NH_2). ^{13}C NMR (DMSO-d_6) δ: 123.5; 128.9; 133.7; 148.1; 151.8; 164.3.

Pyridine-4-carboxylic acid hydrazide (11k) was obtained from 4.4 g (32.1 mmol) **10k**, 4.7 ml (96.3 mmol) of hydrazine hydrate, 3.8 g (86%) of product **11k** was obtained as white crystals.

R_f = 0.40 (10% methanol in chloroform). Mp 172°C. ^{1}H NMR (DMSO-d_6) δ: 4.60 (2H, s, -NH-N$\underline{H}_2$); 7.70-7.72 (2H, m, pyridine ring 3-CH and 5-CH); 8.68-8.70 (1H, m, pyridine ring 2-CH and 6-CH); 10.06 (1H, s, -N$\underline{H}$-NH_2). ^{13}C NMR (DMSO-d_6) δ: 120.9; 140.2; 150.1; 163.8.

General synthetic procedure for ethyl β-*N*-acyloxalamidrazones (12)

Solution of pyridine carbohydrazides **11** and solution 1.1 eq. ethyl 2-thiooxamate each in 15 ml of ethanol were combined. The reaction mass was stirred at room temperature until the full conversion of hydrazide **11** (TLC 10% methanol in chloroform). Then the precipitate was filtered and washed on a filter with 10 ml of ethanol and dried on a filter. The R_f values of the products were given in a system of 15% methanol in chloroform.

Ethyl β-N-(pyridin-2-ylcarbonyl)oxalamidrazone (12i) was obtained from 4.1 g (30.0 mmol) of **11i**, 4.4 g (32.9 mmol) ethyl 2-thiooxamate in 96 hours 6.6 g (93%) of product **12i** was obtained as yellow crystals.

R_f = 0.42. M.p. 192-194 °C. ^{1}H NMR (DMSO-d_6) δ: 1.28 (3H, t, J=7.07, -CH_2-C$\underline{H}_3$); 4.25 (2H, q, J=7.07, -C$\underline{H}_2$-CH_3); 6.89 (2H, s, -NH_2); 7.59-7.61 (1H, m, pyridine ring 5-CH); 7.97-8.04 (2H, m, pyridine ring 3-CH and 4-CH); 8.66-8.67 (1H, m, pyridine ring 6-CH); 10.57 (br.s, -N$\underline{H}$-N=). ^{13}C NMR (DMSO-d_6) δ: 14.0; 61.8; 122.5; 126.7; 137.9; 141.6; 148.4; 149.9; 160.5; 162.2. HRMS: for $C_{10}H_{12}N_4O_3$ $[M+H]^+$ calculated: 237.0987, found: 237.0991.

Ethyl β-N-(pyridin-3-ylcarbonyl)oxalamidrazone (12j) was obtained from 3.5 g (25.5 mmol) of **11j**, 3.7 g (28.1 mmol) ethyl 2-thiooxamate in 96 hours 4.8 g (79%) of product **12j** was obtained as yellow crystals.

R_f = 0.48. M.p. 190-192 °C. ^{1}H NMR (DMSO-d_6) δ: 1.27 (3H, t, J=7.02, -CH_2-C$\underline{H}_3$); 4.25 (2H, q, J=7.02, -C$\underline{H}_2$-CH_3); 6.62-6.81 (2H, m, -NH_2); 7.49-7.53 (1H, m, pyridine ring 5-CH); 8.16-8.19 (1H, m, pyridine ring 4-CH); 8.70-8.71 (1H, m, pyridine ring 6-CH); 8.99 (br.s, pyridine ring 2-CH); 10.21-10.45 (1H, m, -N$\underline{H}$-N=). ^{13}C NMR (DMSO-d_6) δ: 14.0; 61.7; 123.4; 129.8; 135.5; 141.8; 148.6; 151.9; 161.9; 162.1. HRMS: for $C_{10}H_{12}N_4O_3$ $[M+H]^+$ calculated: 237.0987, found: 237.0993.

Ethyl β-N-(pyridin-4-ylcarbonyl)oxalamidrazone (12k) was obtained from 3.7 g (27.4 mmol) of **11k**, 4.0 g (30.0 mmol) ethyl 2-thiooxamate in 96 hours 4.6 g (71%) of product **12k** was obtained as yellow crystals.

R_f = 0.50. M.p. 186-188 °C. ^{1}H NMR (DMSO-d_6) δ: 1.28 (3H, m, -CH_2-C$\underline{H}_3$); 4.26 (2H, m, -C$\underline{H}_2$-CH_3); 6.82 (2H, br.s, -NH_2); 7.74-7.76 (2H, m, pyridine ring 3-CH and 5-CH); 8.71 (2H, br.s, pyridine ring 2-CH and 6-CH); 10.25 (1H, br.s, -N$\underline{H}$-N=). ^{13}C NMR (DMSO-d_6) δ: 14.0; 61.8; 121.7; 141.0; 142.3; 150.0; 161.8; 162.0. HRMS: for $C_{10}H_{12}N_4O_3$ $[M+H]^+$ calculated: 237.0987, found: 237.0994.

General synthetic procedure for ethyl 5-pyridyl-1,2,4-triazole-3-carboxylates (13)

Acyloxalamidrazone **12** was refluxed in *o*-xylene (20 ml of *o*-xylene per 1 g of acyloxalamidrazone) in 24 hours. Then the solvent was evaporated. The reaction product **13** was isolated by column chromatography on silica gel in a chloroform/methanol solvent system (with an methanol gradient from 0 to 5%).

Ethyl 5-(pyridin-2-yl)-1,2,4-triazole-3-carboxylate (13i) was obtained from 6.5 g (27.5 mmol) **12i** in 130 ml of *o*-xylene was obtained 4.5 g (76%) of product **13i** as white crystals.

R_f = 0.40 (5% methanol in chloroform). M.p. 164-166 °C. ^{1}H NMR (DMSO-d_6) δ: 1.32 (3H, t, J=7.07, -CH_2-C$\underline{H}_3$); 4.35 (2H, q, J=7.07, -C$\underline{H}_2$-CH_3); 7.53-7.57 (1H, m, pyridine ring 5-CH); 7.98-8.03 (1H, m, pyridine ring 3-CH); 8.11-8.13 (1H, m, pyridine ring 4-CH); 8.71-8.73 (1H, m, pyridine ring 6-CH). ^{13}C NMR (DMSO-d_6) δ: 14.2; 61.2; 121.7; 125.6; 138.1; 145.8; 149.8; 154.3; 155.7; 159.7. HRMS: for $C_{10}H_{10}N_4O_2$ $[M+H]^+$ calculated: 219.0882, found: 219.0890; $[M-H]^-$ calculated: 217.0725, found: 217.0731.

Ethyl 5-(pyridin-3-yl)-1,2,4-triazole-3-carboxylate (13j) was obtained from 4.7 g (19.9 mmol) **12j** in 100 ml of *o*-xylene was obtained 2.9 g (67%) of product **13j** as white crystals.

R_f = 0.37 (5% methanol in chloroform). M.p. 183-185 °C. ^{1}H NMR (DMSO-d_6) δ: 1.33 (3H, t, J=7.07, -CH_2-C$\underline{H}_3$); 4.37 (2H, q, J=7.07, -C$\underline{H}_2$-CH_3); 7.54-7.58 (1H, m, pyridine ring 5-CH); 8.34-8.37 (1H, m, pyridine ring 4-CH); 8.67-8.69 (1H, m, pyridine ring 6-CH); 9.18 (1H, br.s, pyridine ring 2-CH). ^{13}C NMR (DMSO-d_6) δ: 14.3; 61.6; 124.4; 125.6; 138.1; 145.8; 149.7; 152.3; 154.1; 159.8. HRMS: for $C_{10}H_{10}N_4O_2$ $[M+H]^+$ calculated: 219.0882, found: 219.0892; $[M-H]^-$ calculated: 217.0725, found: 217.0730.

Ethyl 5-(pyridin-4-yl)-1,2,4-triazole-3-carboxylate (13k) was obtained from 4.5 g (19.0 mmol) **12k** in 90 ml of *o*-xylene was obtained 3.3 g (80%) of product **13k** as white crystals.

R_f = 0.53 (5% methanol in chloroform). M.p. 177-179 °C. ^{1}H NMR (DMSO-d_6) δ: 1.34 (3H, t, J=7.07, -CH_2-C$\underline{H}_3$); 4.38 (2H, t, J=7.07, -C$\underline{H}_2$-CH_3); 7.94-7.96 (2H, m, pyridine ring 3-CH and 5-CH); 8.72-8.74 (2H, br.s, pyridine ring 2-CH and 6-CH). ^{13}C NMR (DMSO-d_6) δ: 14.0; 61.6; 120.2; 135.5; 150.6; 152.6; 156.9; 158.2. HRMS: for $C_{10}H_{10}N_4O_2$ $[M+H]^+$ calculated: 219.0882, found: 219.0887; $[M-H]^-$ calculated: 217.0725, found: 217.0729.

General synthetic procedure for 5-pyridyl-1,2,4-triazole-3-carboxamides (4)

An ammonia solution in anhydrous methanol (12 M) was added to ethyl 5-pyridyl-1,2,4-triazole-3-carboxylate **13** (10 ml per 1g of **9**)б and the mixture was stirred at the reflux for 24 hours and 1 ml of ammonia solution in anhydrous methanol (12 M) was added every two hours. Then the volatile components were evaporated. The product **4** was isolated by column chromatography on silica gel in a methanol-chloroform solvent system (with an methanol gradient from 0 to 25%).

5-(Pyridin-2-yl)-1,2,4-triazole-3-carboxamide (4i) was obtained from 0.50 g (2.29 mmol) of ethyl 5-(pyridin-2-yl)-1,2,4-triazole-3-carboxylate **13i**, 0.29 g (68%) of the product **4i** was obtained, as white crystals.

R_f = 0.20 (15% methanol in chloroform). M.p. >300 °C. ^{1}H NMR (DMSO-d_6) δ: 7.55 (1H, br.s, pyridine ring 5-CH); 7.60 and 7.86 (2H, 2 br.s, -CON$\underline{H}_2$); 8.02-8.12 (2H, m, pyridine ring 3-CH and 4-CH); 8.71 (1H, br.s, pyridine ring 6-CH). ^{13}C NMR (DMSO-d_6+DCl) δ: 121.7; 125.3; 138.4; 146.3; 149.1; 155.6; 156.1; 160.0. HRMS: for $C_8H_7N_5O$ $[M+H]^+$ calculated: 190.0729, found: 190.0735; $[M-H]^-$ calculated: 188.0572, found: 188.0579.

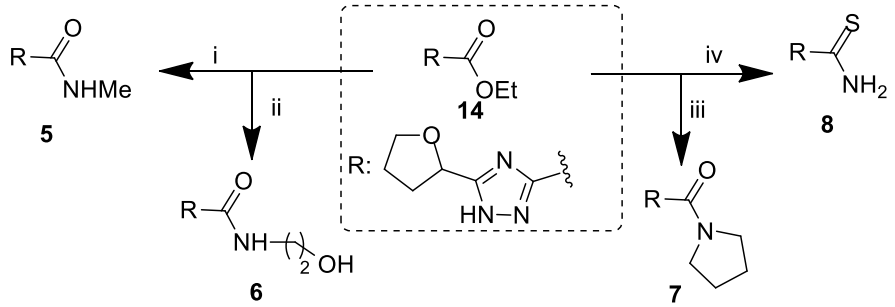
5-(Pyridin-3-yl)-1,2,4-triazole-3-carboxamide (4j) was obtained from 0.50 g (2.29 mmol) of ethyl 5-(pyridin-3-yl)-1,2,4-triazole-3-carboxylate **13j**, 0.37 g (85%) of the product **4j** was obtained, as white crystals.

R_f = 0.25 (10% methanol in chloroform). M.p. 295-297 °C. ^{1}H NMR (DMSO-d_6) δ: 7.51-7.56 (1H, m, pyridine ring 5-CH); 7.88 and 8.12 (2H, 2 br.s, -CONH$_2$); 8.33-8.35 (1H, m, pyridine ring 4-CH); 8.64-8.65 (1H, m, pyridine ring 6-CH); 9.20 (1H, br.s, pyridine ring 2-CH). ^{13}C NMR (DMSO-d_6+DCl) δ: 124.1; 125.9; 133.5; 146.7; 148.1; 150.5; 151.9; 158.9. HRMS: for $C_8H_7N_5O$ $[M+H]^+$ calculated: 190.0729, found: 190.0737; $[M-H]^-$ calculated: 188.0572, found: 188.0578.

5-(Pyridin-4-yl)-1,2,4-triazole-3-carboxamide (4k) was obtained from 0.50 g (2.29 mmol) of ethyl 5-(pyridin-4-yl)-1,2,4-triazole-3-carboxylate **13k**, 0.39 g (89%) of the product **4k** was obtained, as white crystals.

R_f = 0.30 (12% methanol in chloroform). M.p. >300 °C. ^{1}H NMR (DMSO-d_6) δ: 7.89 and 8.10 (2H, 2 br.s, -CON$\underline{H}_2$); 7.93-7.95 (2H, m, - pyridine ring 3-CH and 5-CH); 8.70-8.72 (2H, br.s, pyridine ring 2-CH and 6-CH). ^{13}C NMR (DMSO-d_6+DCl) δ: 121.7; 132.8; 146.2; 152.3; 155.8; 158.2. HRMS: for $C_8H_7N_5O$ $[M+H]^+$ calculated: 190.0729, found: 190.0739; $[M-H]^-$ calculated: 188.0572, found: 188.0576.

Synthesis of 5-(tetrahydrofuran-2-yl)-1,2,4-triazole-3-carboxamides modified at position 3



Scheme S3 *Reagents and conditions:* i, 10M $MeNH_2$ in MeOH (10 eq), 12 h, r.t.; ii, $NH_2CH_2CH_2OH$ (2 eq), EtOH, 12 h, r.t.; iii, 1. NaOH (2.2 eq), EtOH, 24 h, r.t; 2. $SOCl_2$ (10 eq), CH_2Cl_2, 48 h, r.t.; 3. pyrrolidine (4 eq), 24 h, r.t; iv, 1. NH_3 in MeOH, 48 h, reflux; 2. P_4S_{10} (0.25 eq), toluene, 2 h, reflux.

Ethyl 5-tetrahydrofuran-2yl-1,2,4-triazole-3-carboxylate (14) was synthesized as previously described.[S2]

N-Methyl-5-(tetrahydrofuran-2-yl)-1,2,4-triazole-3-carboxamide (5).

Ethyl 5-(tetrahydrofuran-2-yl)-1,2,4-triazole-3-carboxylate **14** (0.20 g, 0.95 mmol) was dissolved in a 10-fold mol. excess of methylamine solution (10M) in methanol. The reaction mixture was stirred for 12 hours and then evaporated. The substance was isolated using column chromatography, eluent – chloroform. Yield: 90 mg (48%), product was obtained as white crystals.

R_f = 0.30 (5% methanol in chloroform). M.p. 127-129 °C. ^{1}H NMR ($CDCl_3$): 1.95 -2.06 (2H, m, $OCH_2CH_2C\underline{H_2}$); 2.16-2.41 (2H, m, $OCH_2C\underline{H_2}CH_2$); 3.01 (3H, d, J = 4.95, -NH-C$\underline{H_3}$); 3.88-4.08 (2H, m, -O-CH_2-); 5.14 (1H, t, J=6.72, -O-CH); 7.54 (1H, br. s., -N$\underline{H}$-CH_3). ^{13}C NMR ($CDCl_3$): 25.6; 26.3; 31.7; 69.3; 73.4; 152.6; 158.6; 161.3. HRMS: for $C_8H_{12}N_4O_2$ $[M+H]^+$ calculated: 197.1039, found: 197.1045.

N-(2-Hydroxyethyl)-5-(tetrahydrofuran-2-yl)-1,2,4-triazole-3-carboxamide (6)

Ethyl 5-(tetrahydrofuran-2-yl)-1,2,4-triazole-3-carboxylate **14** (0.20 g, 0.95 mmol) was dissolved in 1 ml of ethanol and 0.12 ml (2 mmol) of ethanolamine was added. The reaction mass was stirred for 12 hours and then evaporated. The product was isolated using column chromatography on silica gel, eluent chloroform with a methanol gradient from 0 to 2%. Yield: 139 mg (65%), product was obtained as yellow oil.

R_f = 0.20 (5% methanol in chloroform).^{1}H NMR (DMSO-d_6): 1.89-2.30 (4H, m, -O-CH_2-C$\underline{H_2}$-C$\underline{H_2}$-); 3.27-3.50 (4H, m, -C$\underline{H_2}$-C$\underline{H_2}$-OH); 3.75-3.94 (2H, m, -O-C$\underline{H_2}$-CH_2-CH_2-); 4.76 (1H, br.s., -OH); 4.98 (1H, t, J = 6.72, -O-C$\underline{H}$); 8.35 (1H, br.s., -N$\underline{H}$-CH_2-). ^{13}C NMR (DMSO-d_6 and CF_3COOH): 25.6; 31.3; 41.8; 59.8; 68.4; 73.1; 154.1; 158.4; 160.7. HRMS: for $C_9H_{14}N_4O_3$ $[M+H]^+$ calculated: 227.1144, found: 227.1148.

3-(Pyrrolidin-1-ylcarbonyl)-5-(tetrahydrofuran-2-yl)-1,2,4-triazole (7)

To 0.30 g (5.3 mmol) of sodium hydroxide in 2 ml of ethanol, 0.5 g (2.4 mmol) of ethyl 5-(tetrahydrofuran-2-yl)-1,2,4-triazole-3-carboxylate **14** was added. The reaction was stirred for 24 hours at room temperature and evaporated. To the residue 1.74 ml (24 mmol) of $SOCl_2$ were added and stirred at room temperature for 24 hours, then evaporated. The residue was suspended in 5 ml of absolute dichloromethande, and 0.8 ml (9.6 mmol) of pyrrolidine was added while cooling to 0°C and stirred for 2 hours. Then the reaction mass was evaporated. To the residue 3 ml of a 1M hydrochloric acid solution was added and extracted with chloroform3 (3 x 5 ml). The combined organic phases were dried, filtered off and the solvent was evaporated and the product was isolated by column chromatography on silica gel, eluent chloroform with a methanol gradient from 0 to 10%. Yield: 0.15 g (26%), product was obtained as yellow oil.

R_f = 0.58 (5% methanol in chloroform).^{1}H NMR ($CDCl_3$): 1.86-2.39 (8H, m, -O-CH_2-C$\underline{H_2}$-C$\underline{H_2}$- and -N-CH_2-C$\underline{H_2}$-C$\underline{H_2}$-); 3.69 (2H, t, J=6.72, -CH_2-O-); 3.89-4.10 (4H, m, -CH_2-N-CH_2-); 5.12 (1H, t, J= 6.72, -O-CH). ^{13}C NMR ($CDCl_3$): 23.7; 25.8; 26.4; 31.2; 47.2; 49.0; 68.9; 74.2; 152.1; 156.9; 163.3. HRMS: for $C_{11}H_{16}N_4O_2$ $[M+H]^+$ calculated: 237.1351, found: 237.1354.

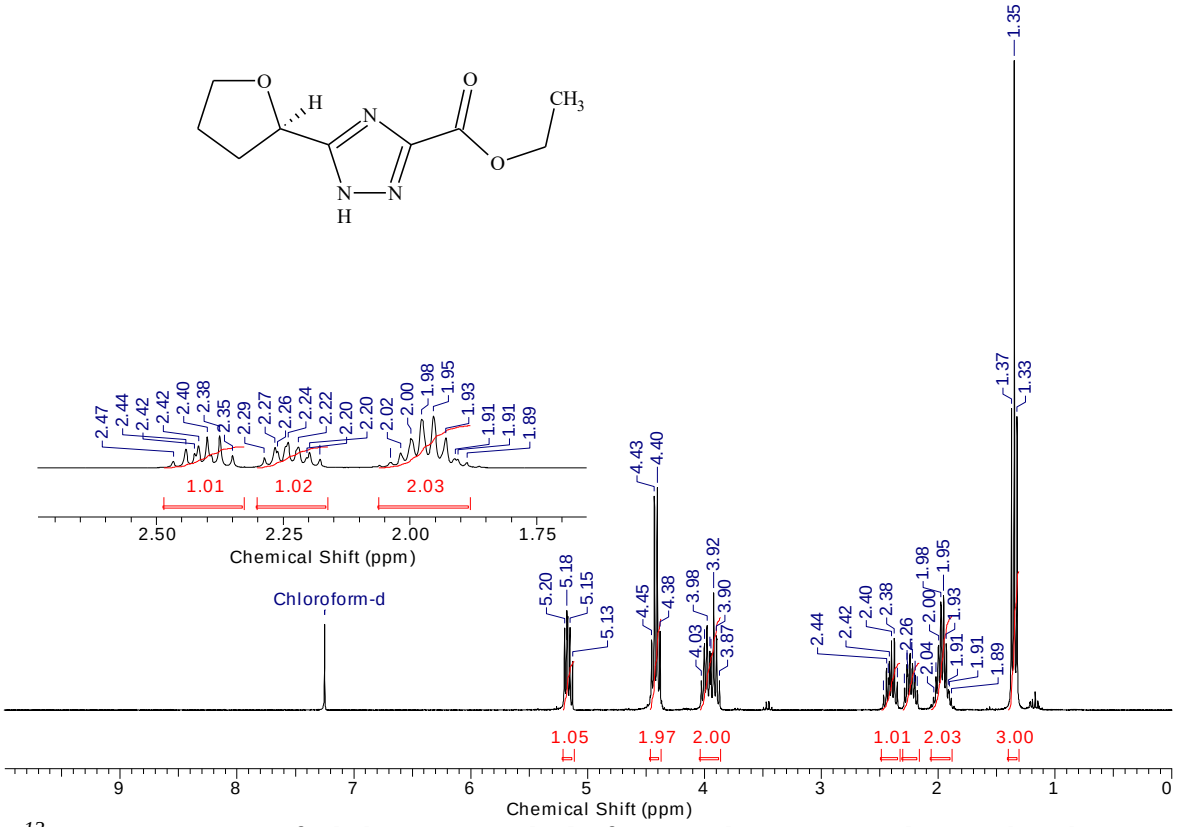
5-(Tetrahydrofuran-2-yl)-1,2,4-triazole-3-carbothioamide (8)

To a suspension of phosphorus(V) sulfide 0.10 g (0.45 mmol) in 2 ml of absolute toluene 0.26 g (1.43 mmol) 5-(tetrahydrofuran-2-yl)-1,2,4-triazole-3-carboxamide was added with constant stirring. The reaction was reflux for 2 hours. Then the hot mass was decanted from the oily residue, the solvent was evaporated. The product was isolated using silica gel column chromatography, eluent chloroform with a methanol gradient from 0 to 10%. Yield: 90 mg (29%), product was obtained as white crystals.

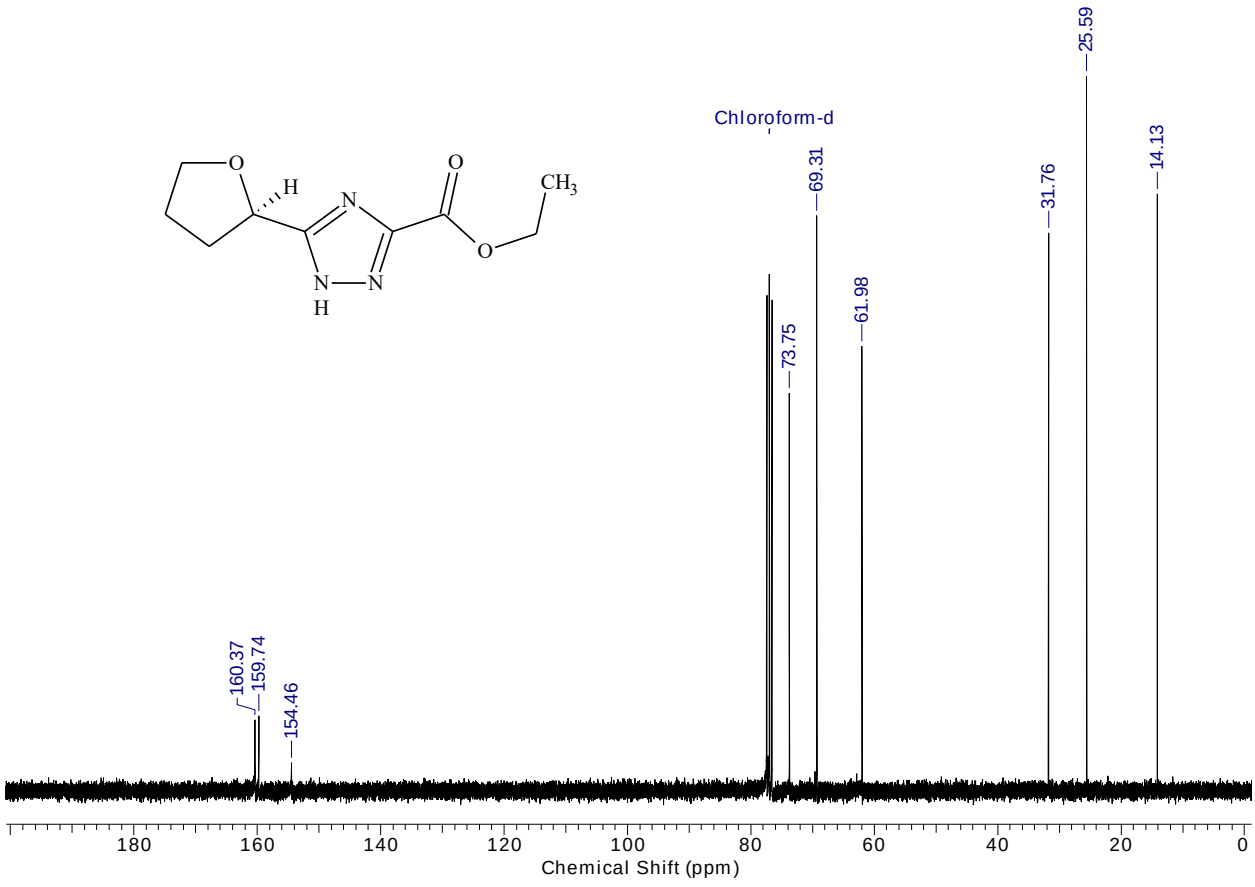
R_f = 0.46 (5% methanol in chloroform). M.p. 178-180 °C. ^{1}H NMR (DMSO-d_6): 1.93-2.24 (4H, m, -O-CH_2-C$\underline{H_2}$-C$\underline{H_2}$-); 3.80-3.90 (2H, m, -O-C$\underline{H_2}$-CH_2-CH_2-); 4.99 (1H, br.s, -O-C$\underline{H}$); 9.36-10.11 (2H, m, -N$\underline{H_2}$). ^{13}C NMR (DMSO-d_6): 25.4; 31.0; 68.1; 73.1; 158.1; 160.8; 184.0. HRMS: for $C_7H_{10}N_4OS$ $[M+H]^+$ calculated: 199.0654, found: 199.0659.

NMR Spectra

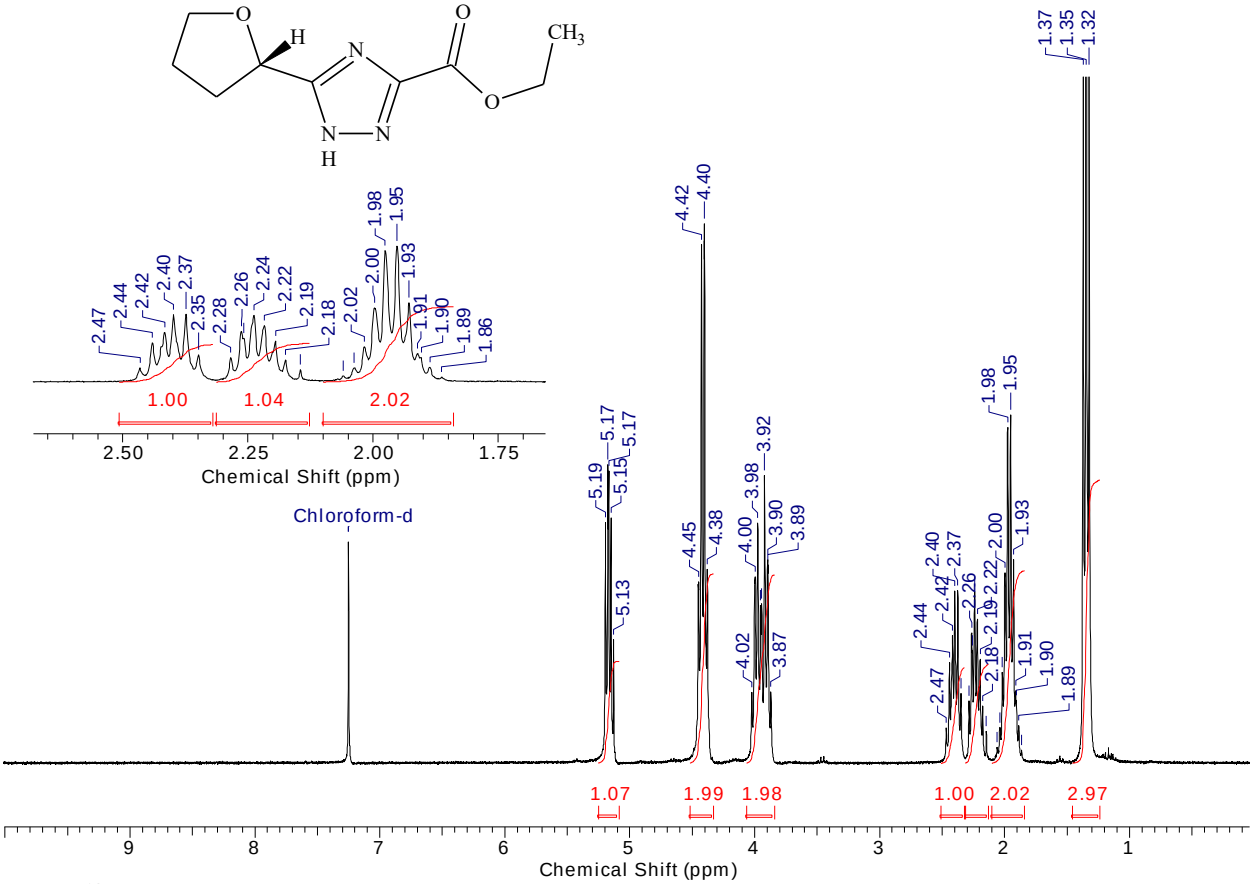
^{1}H NMR spectrum of ethyl 5-((R)-tetrahydrofuran-2-yl)-1,2,4-triazole-3-carboxylate



^{13}C NMR spectrum of ethyl 5-((R)-tetrahydrofuran-2-yl)-1,2,4-triazole-3-carboxylate



^{1}H NMR spectrum of ethyl 5-((S)-tetrahydrofuran-2-yl)-1,2,4-triazole-3-carboxylate



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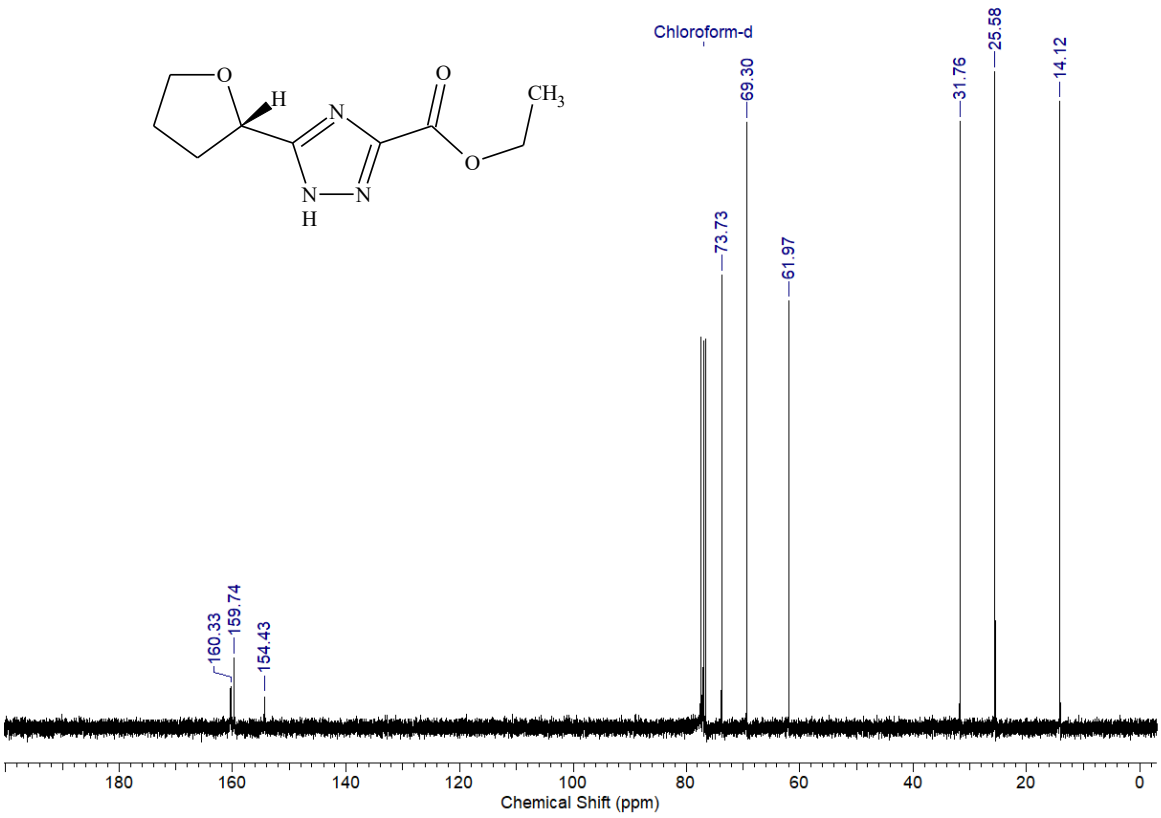
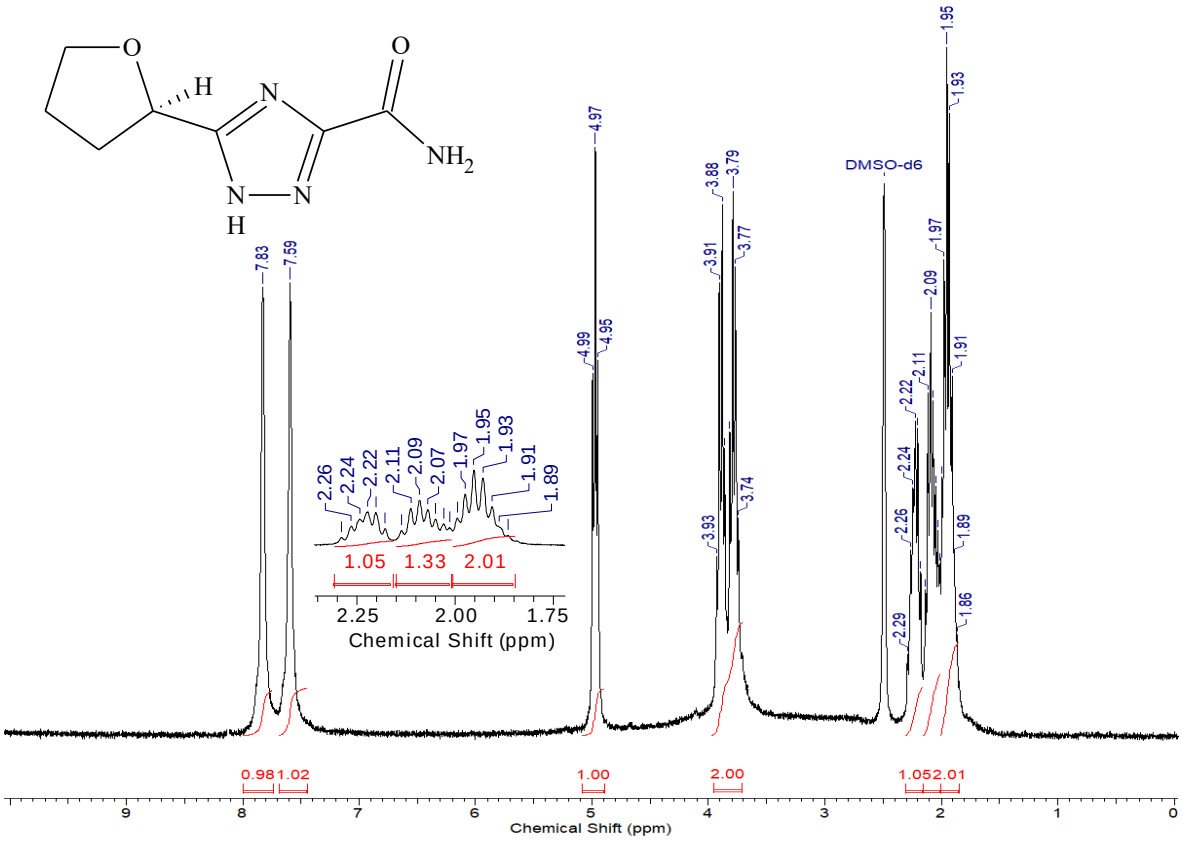
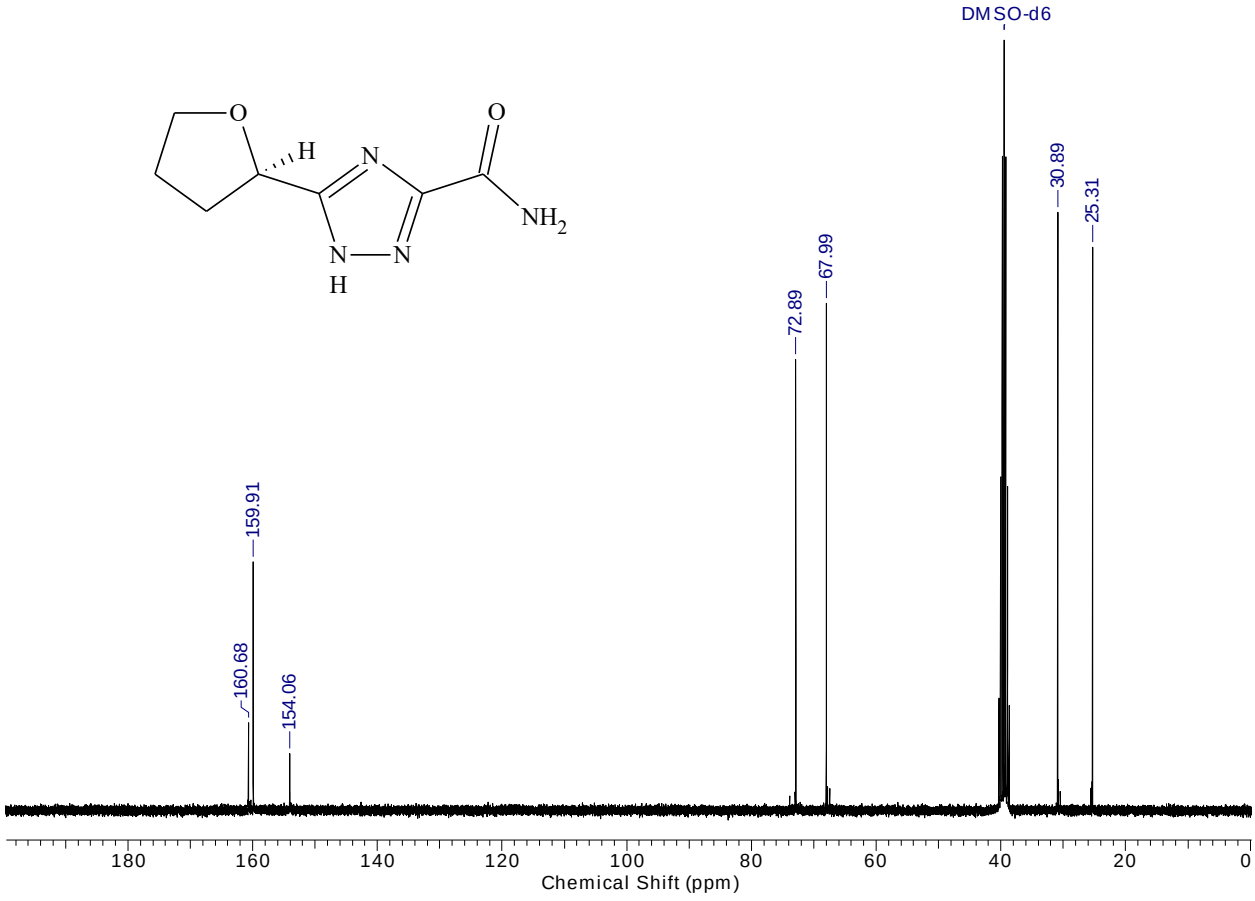


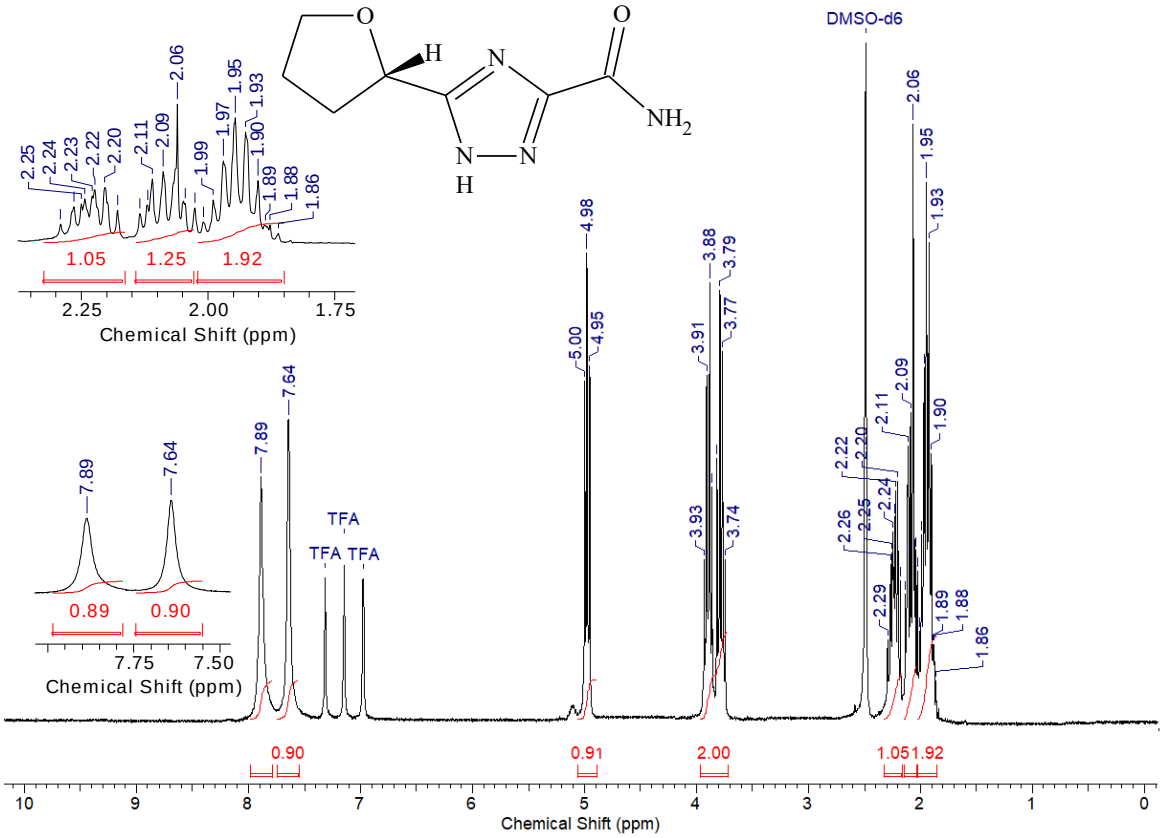
*^{1}H NMR spectrum of **2a***



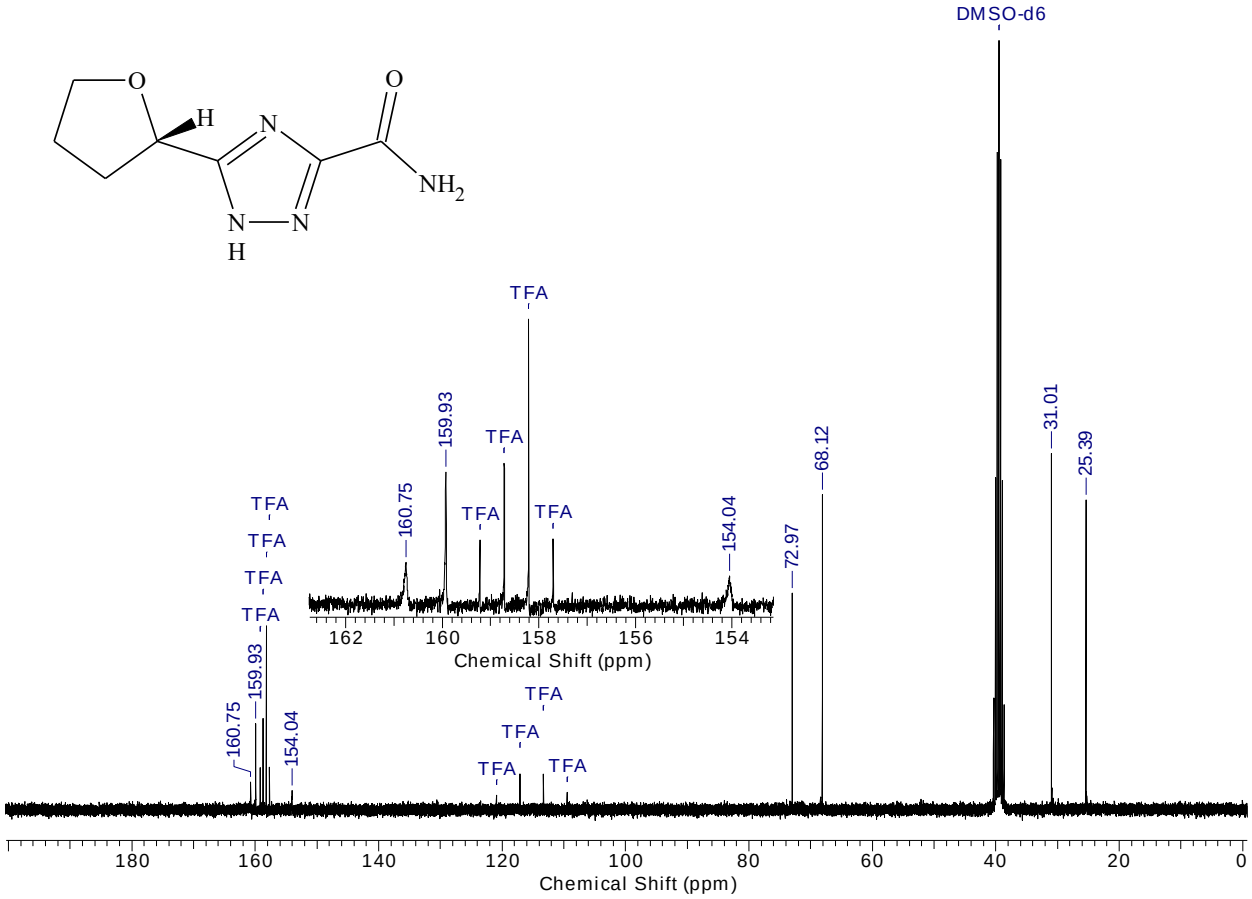
*^{13}C NMR spectrum of **2a***



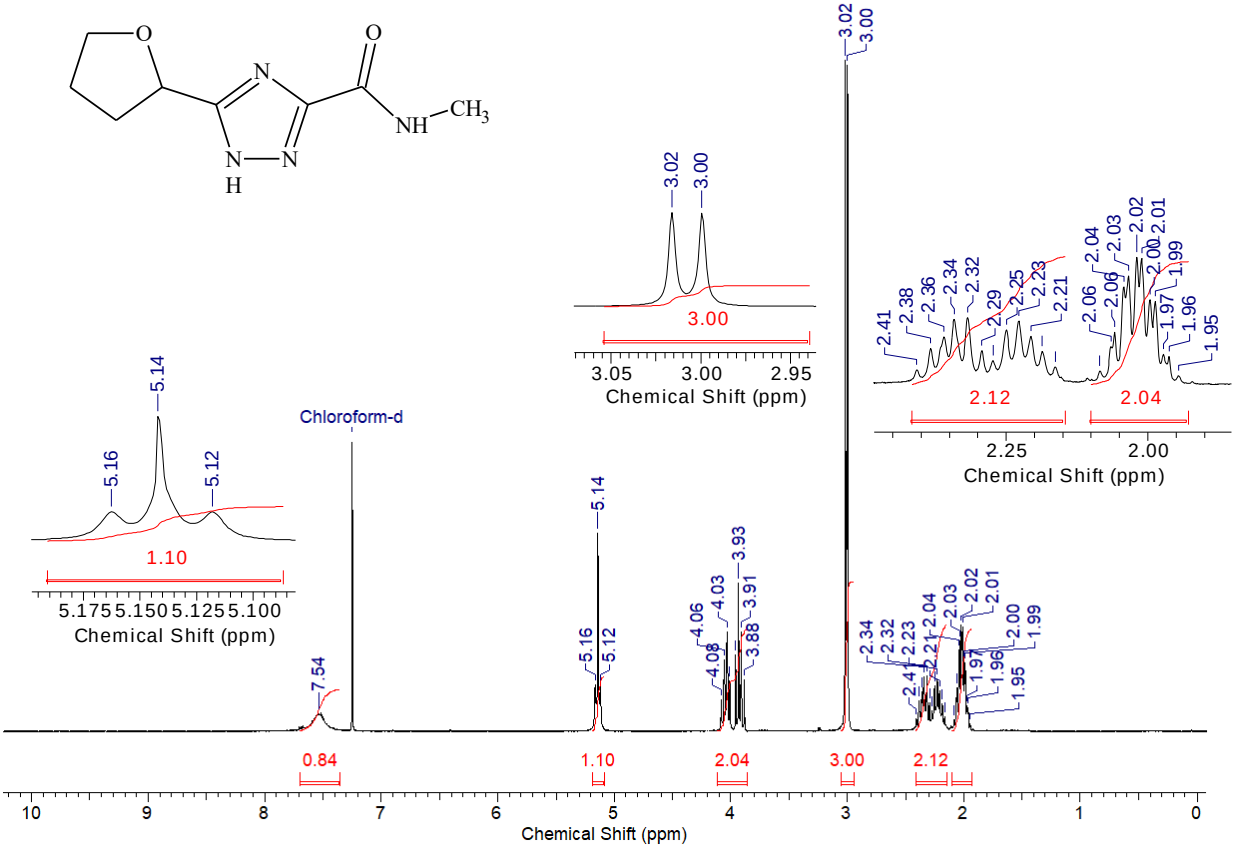
*^{1}H NMR spectrum of **2b***



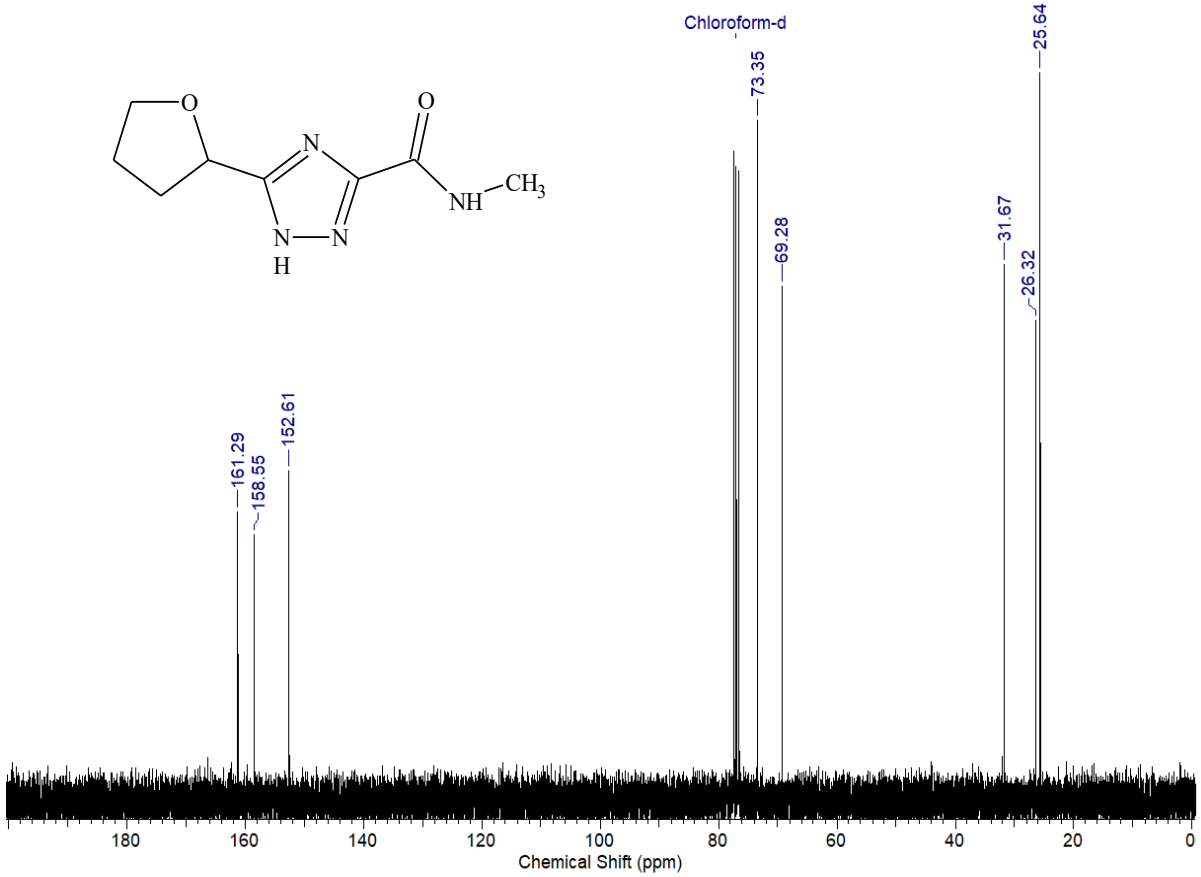
*^{13}C NMR spectrum of **2b***



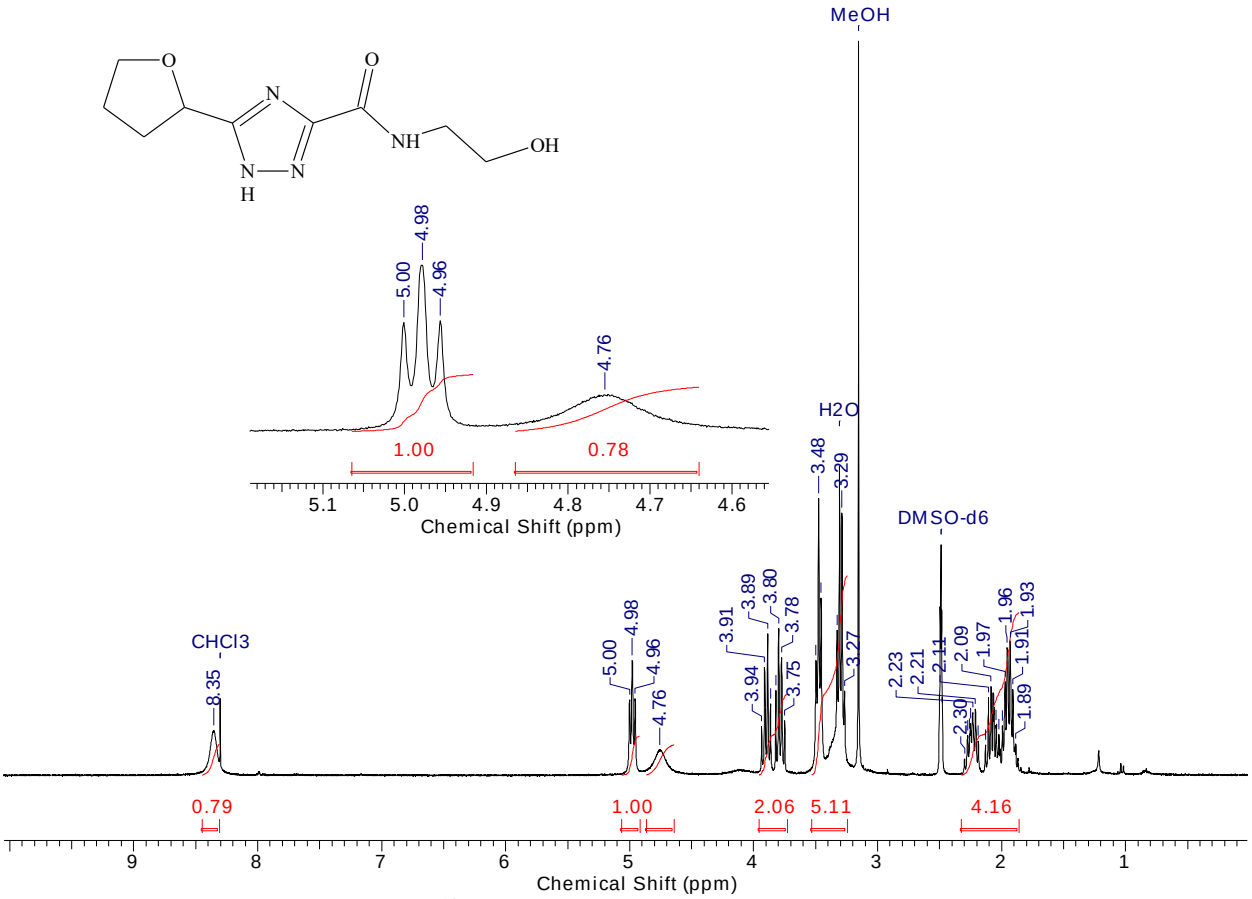
*^{1}H NMR spectrum of **5***



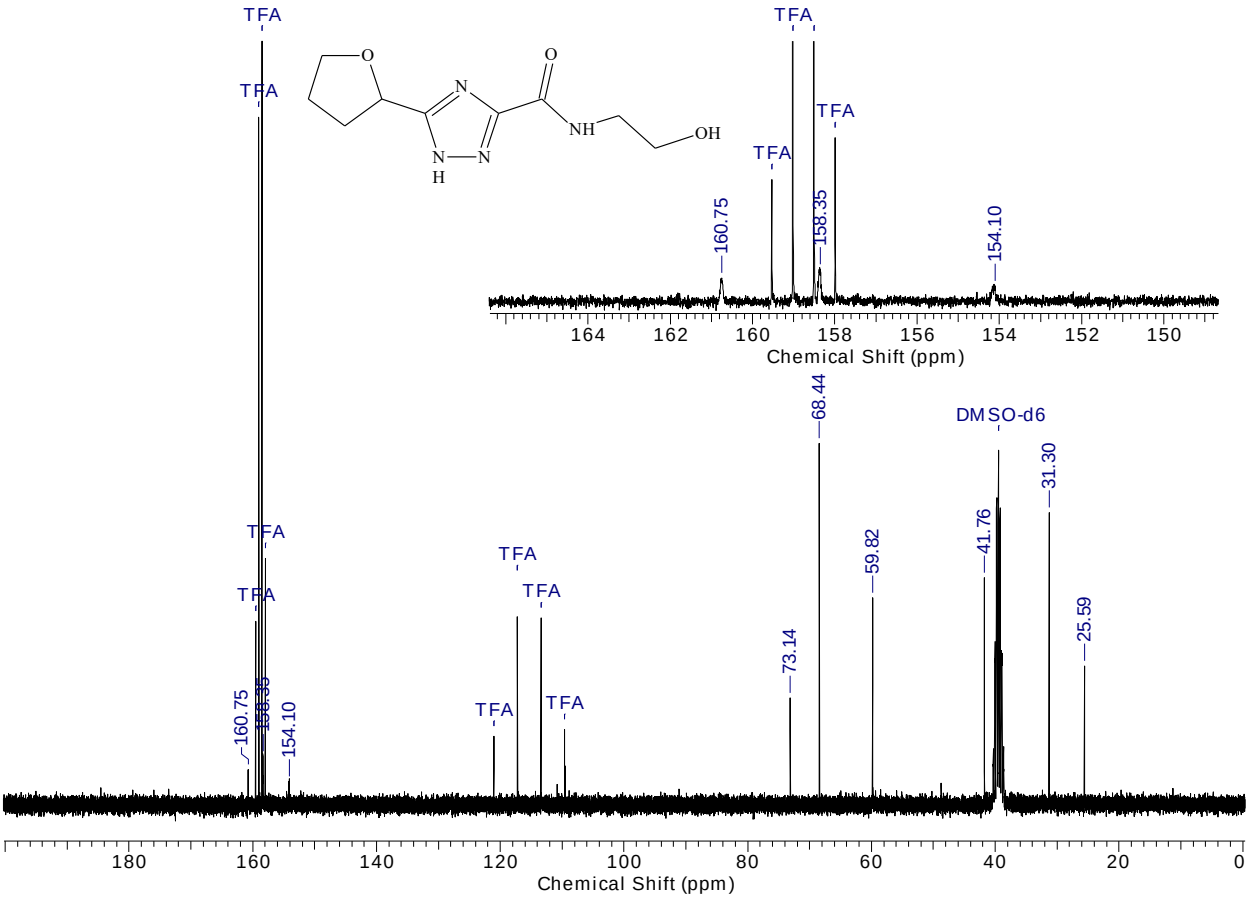
*^{13}C NMR spectrum of **5***



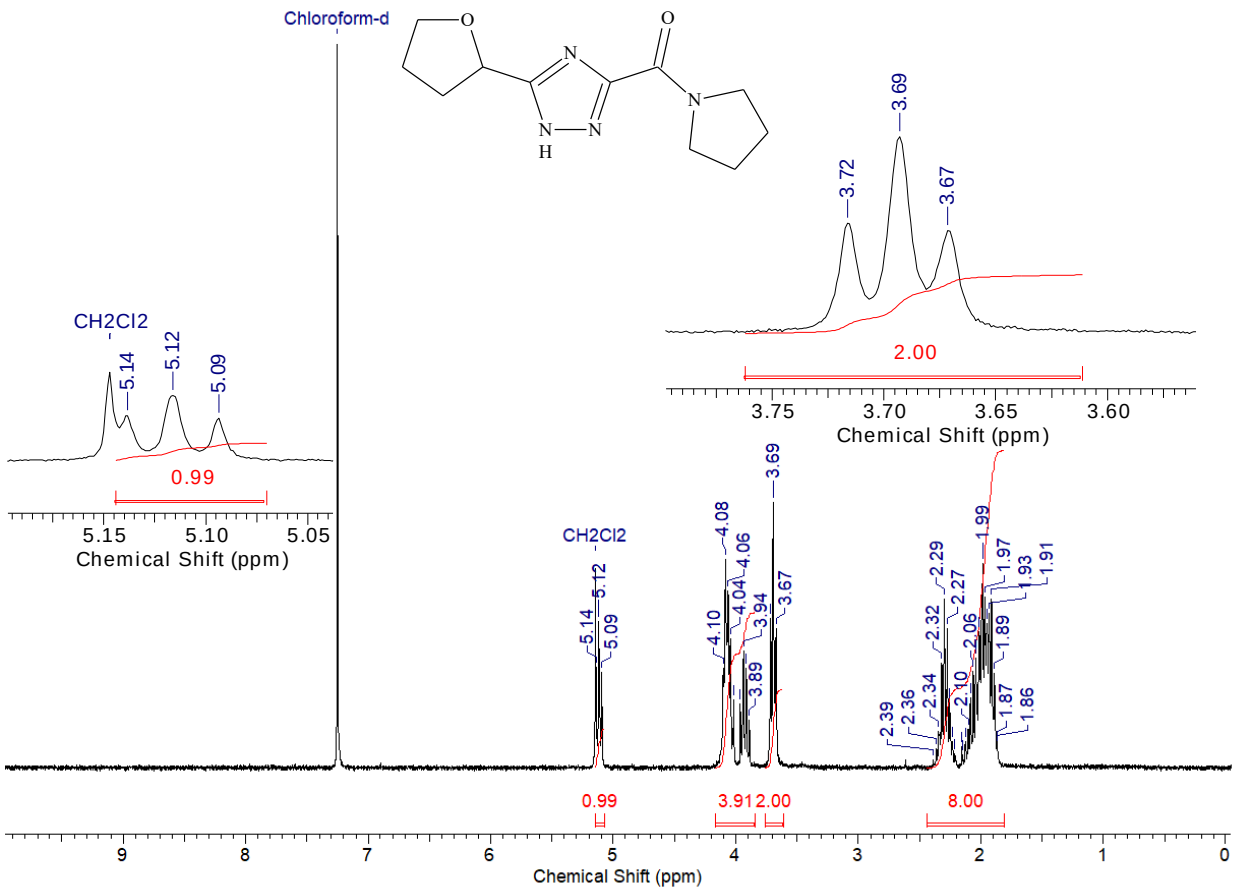
^{1}H NMR spectrum of ***6***



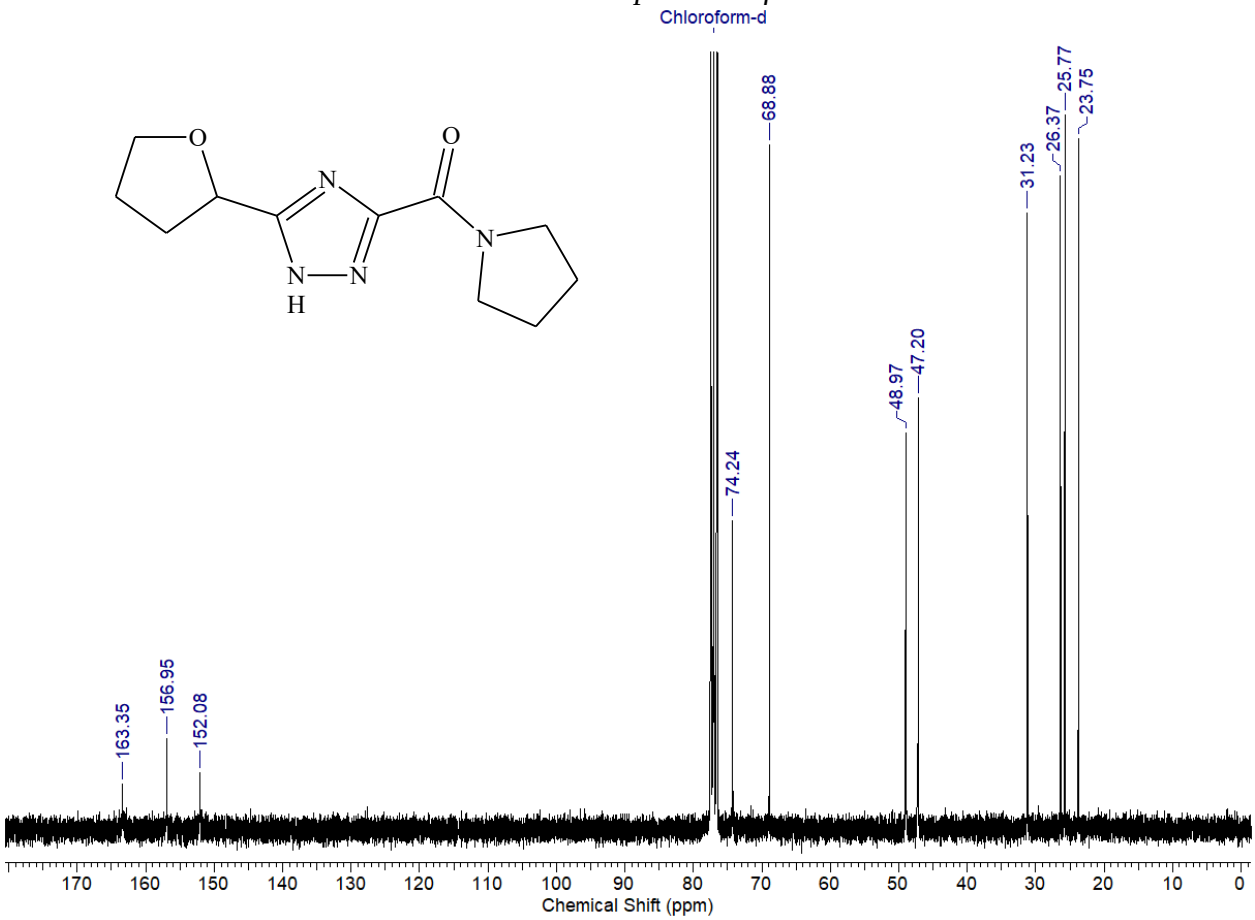
^{13}C NMR spectrum of ***6***



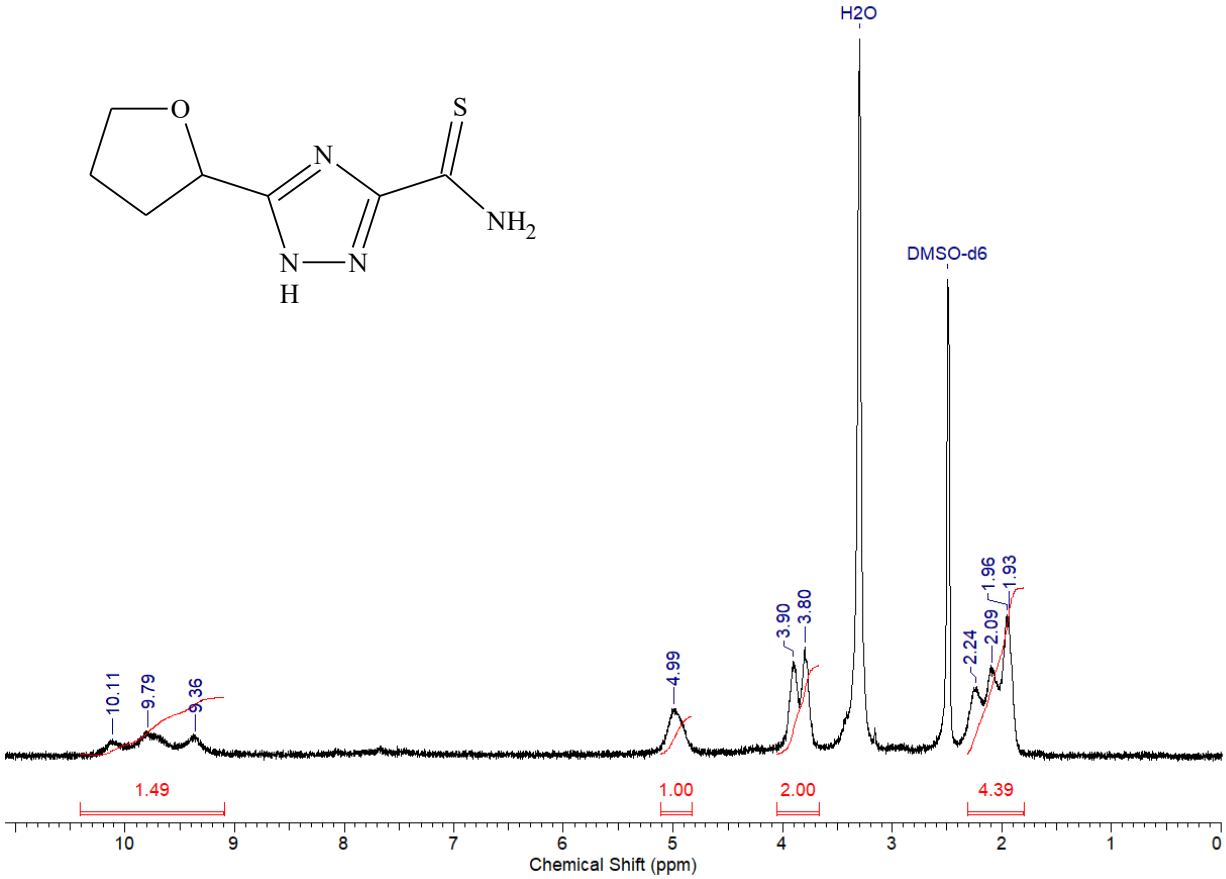
^{1}H NMR spectrum of 7



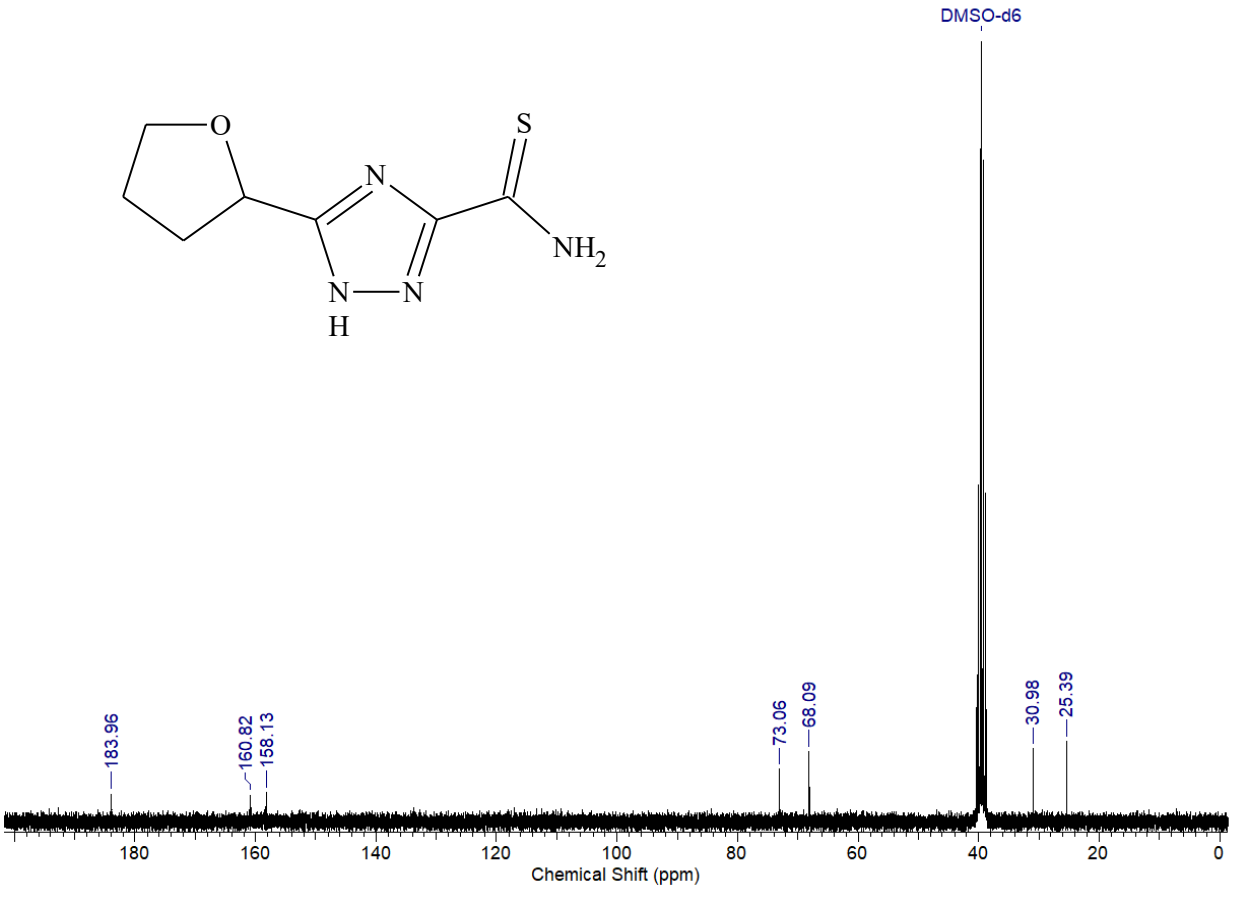
^{13}C NMR spectrum of 7



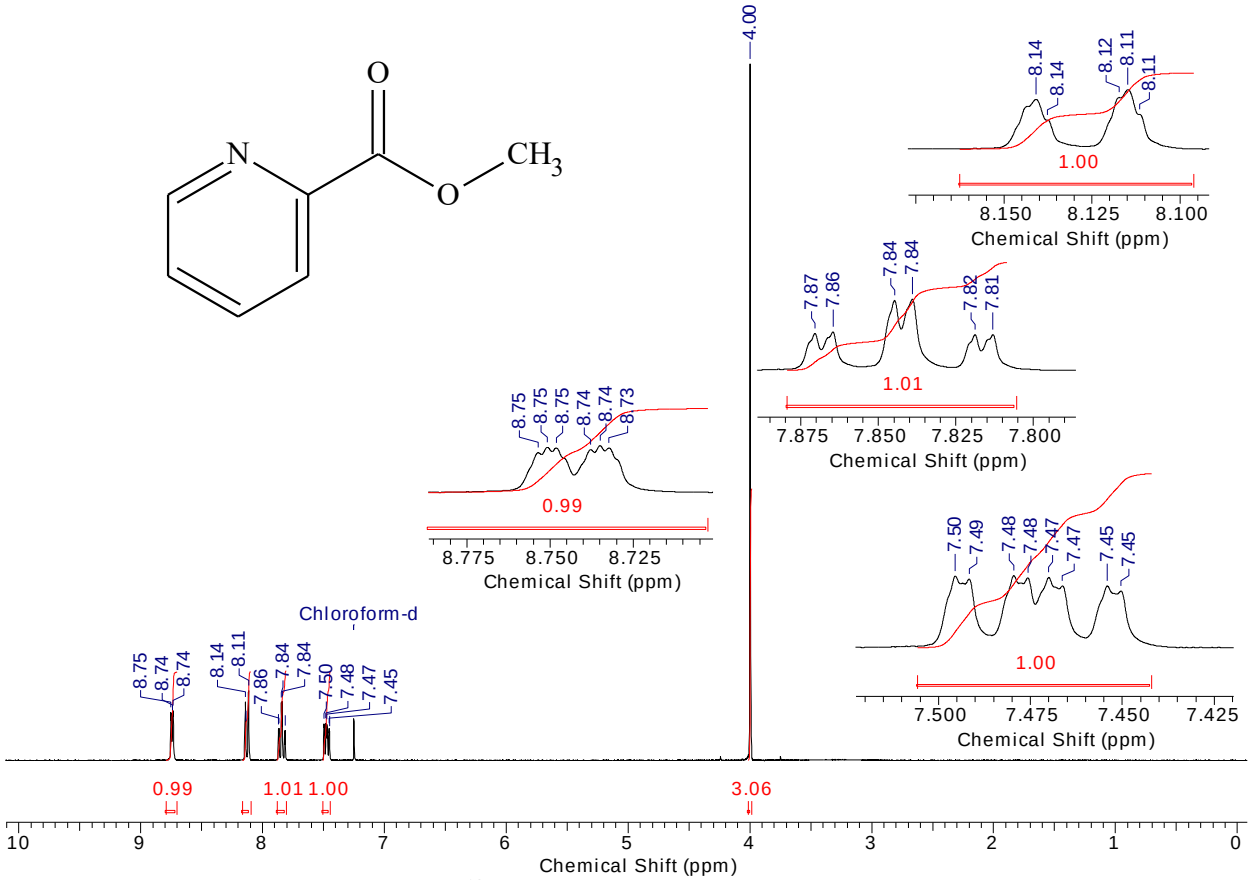
*^{1}H NMR spectrum of **8***



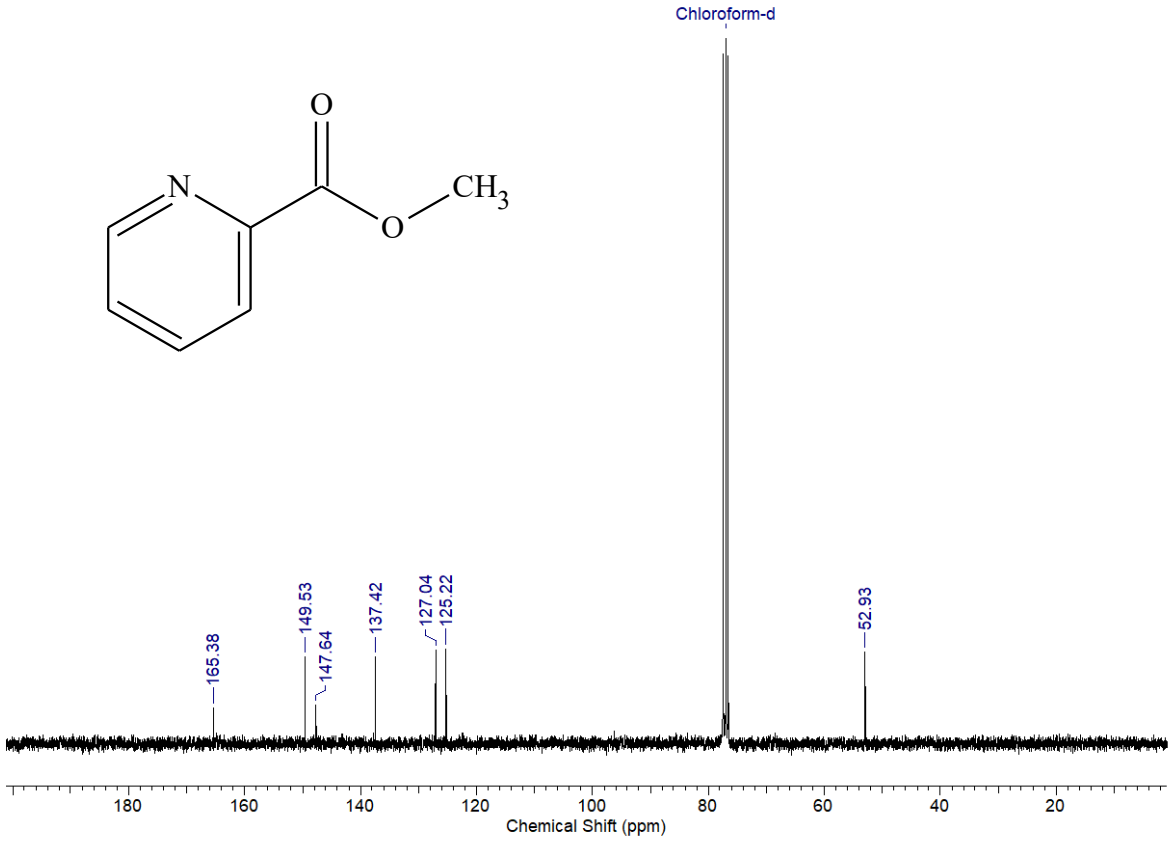
*^{13}C NMR spectrum of **8***



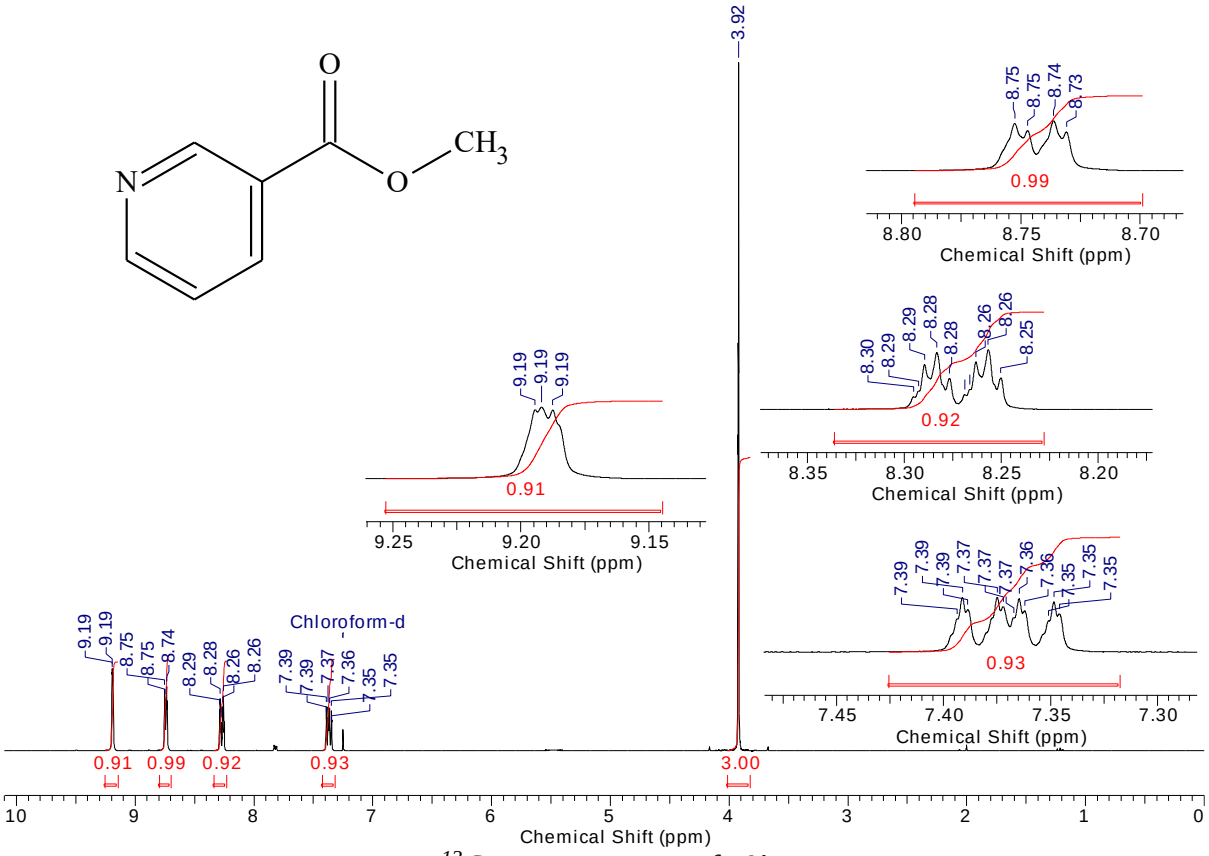
*^{1}H NMR spectrum of **10i***



*^{13}C NMR spectrum of **10i***



*^{1}H NMR spectrum of **10j***



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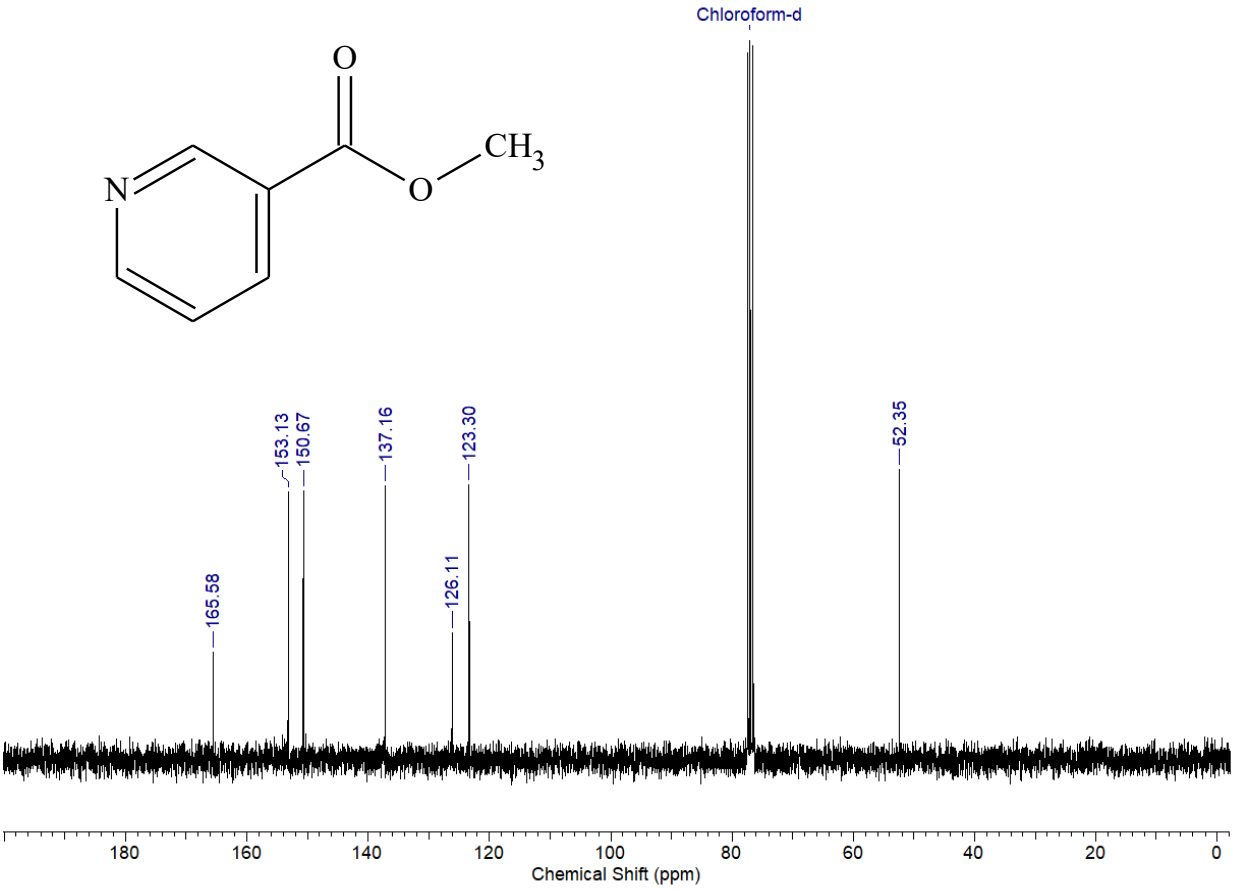

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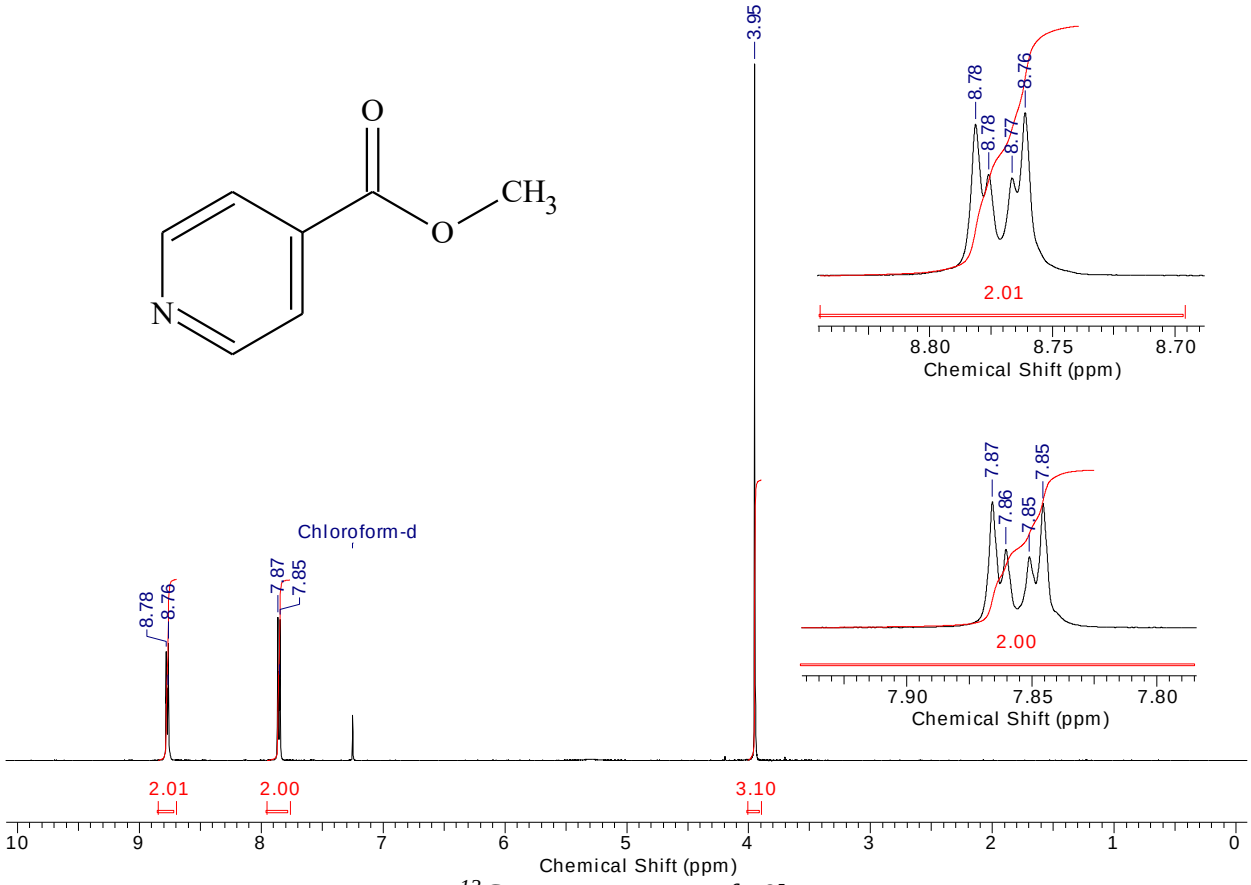

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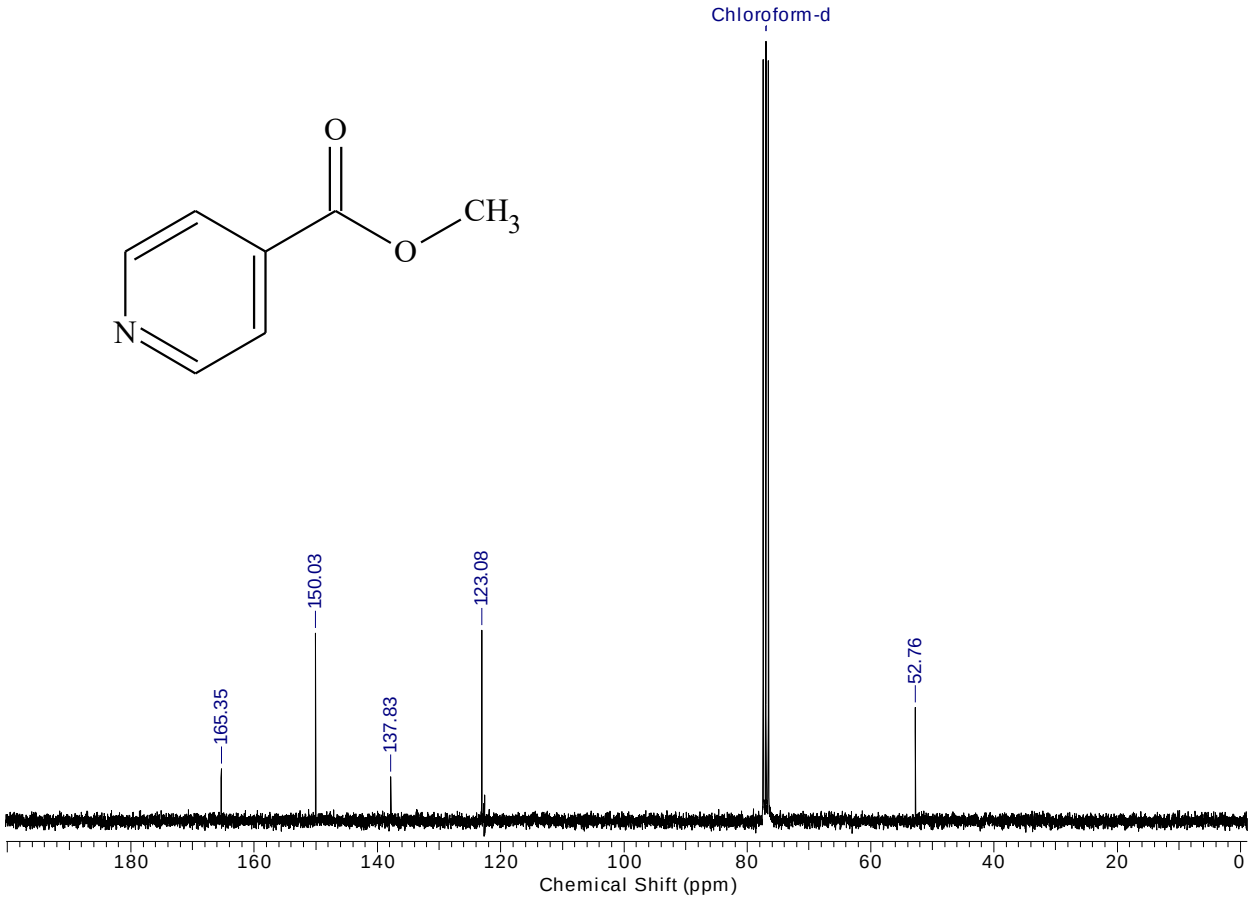
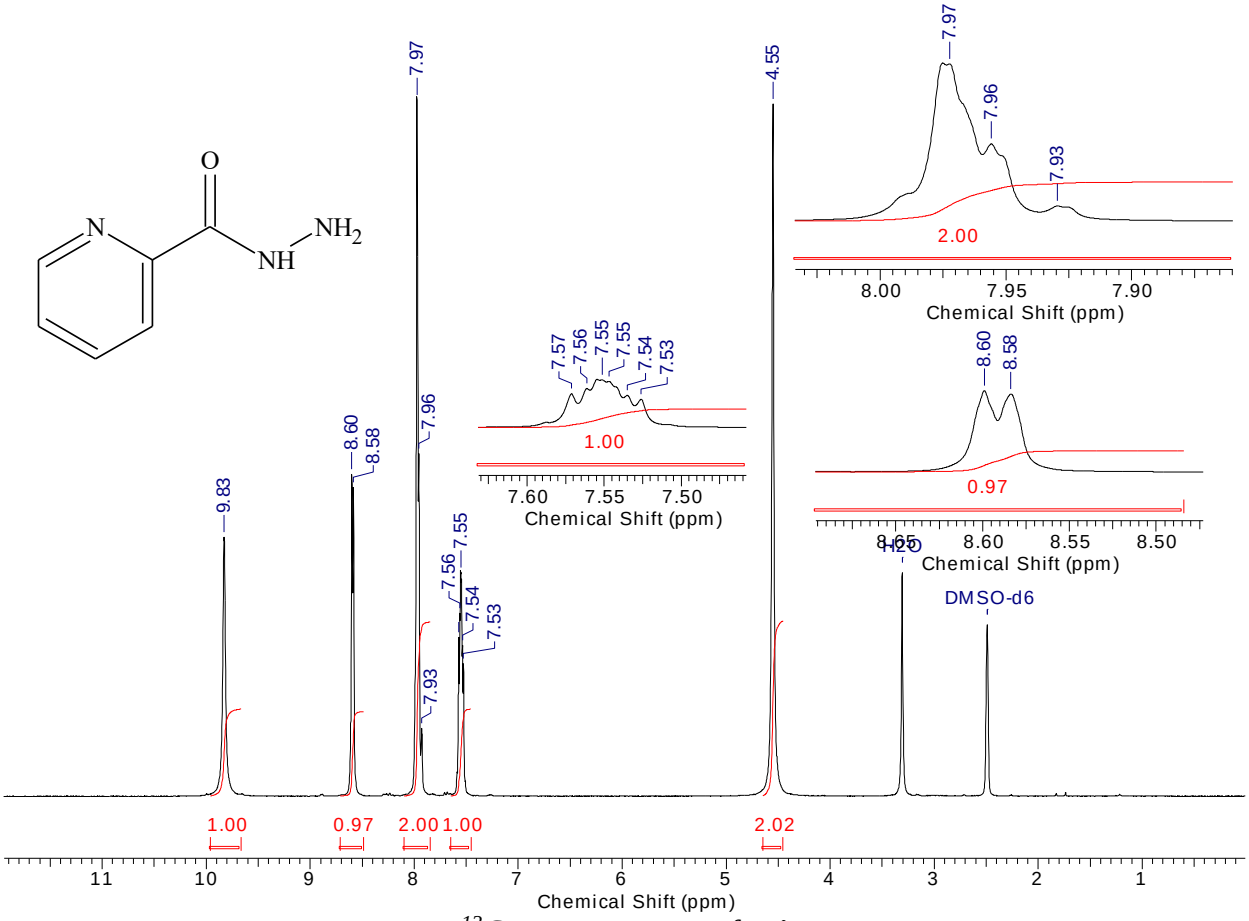
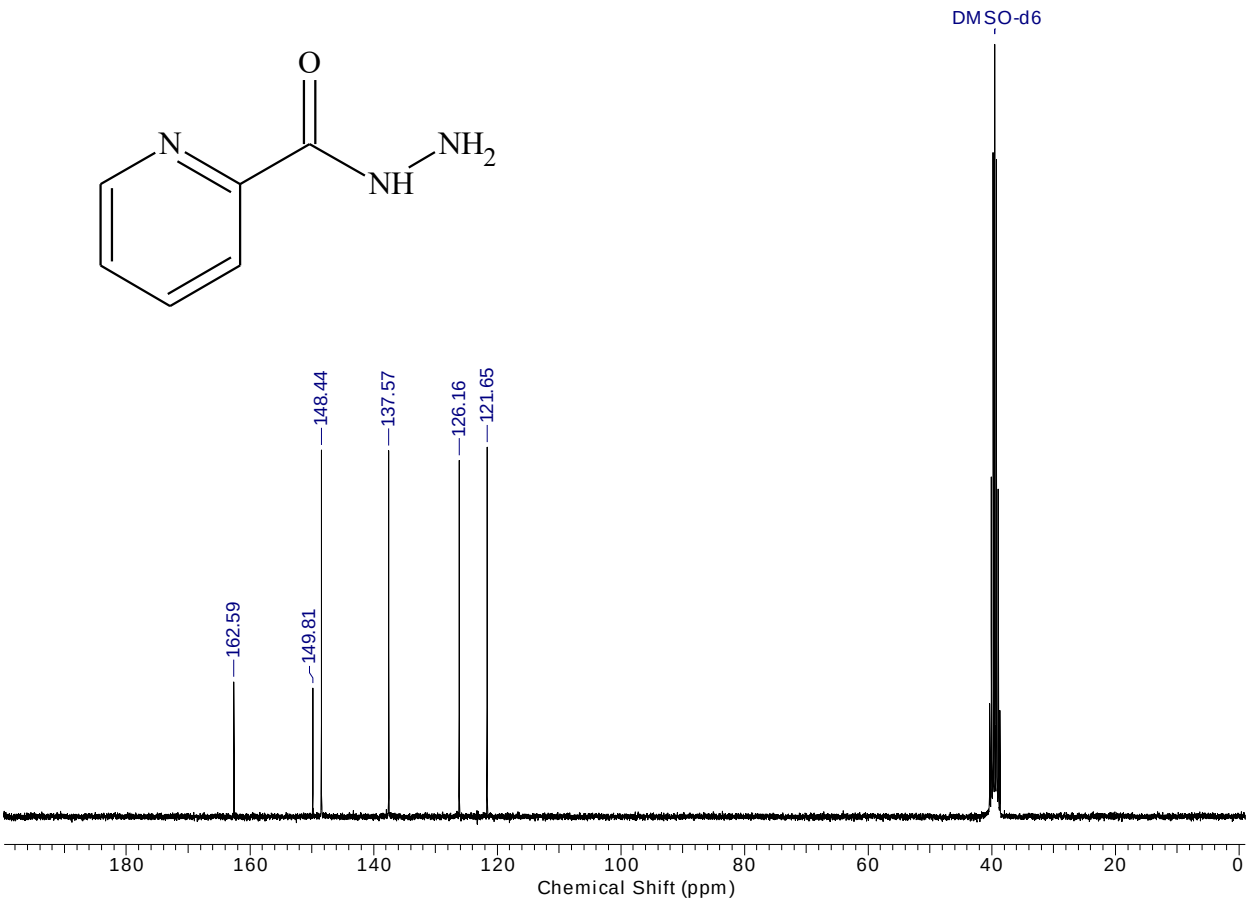


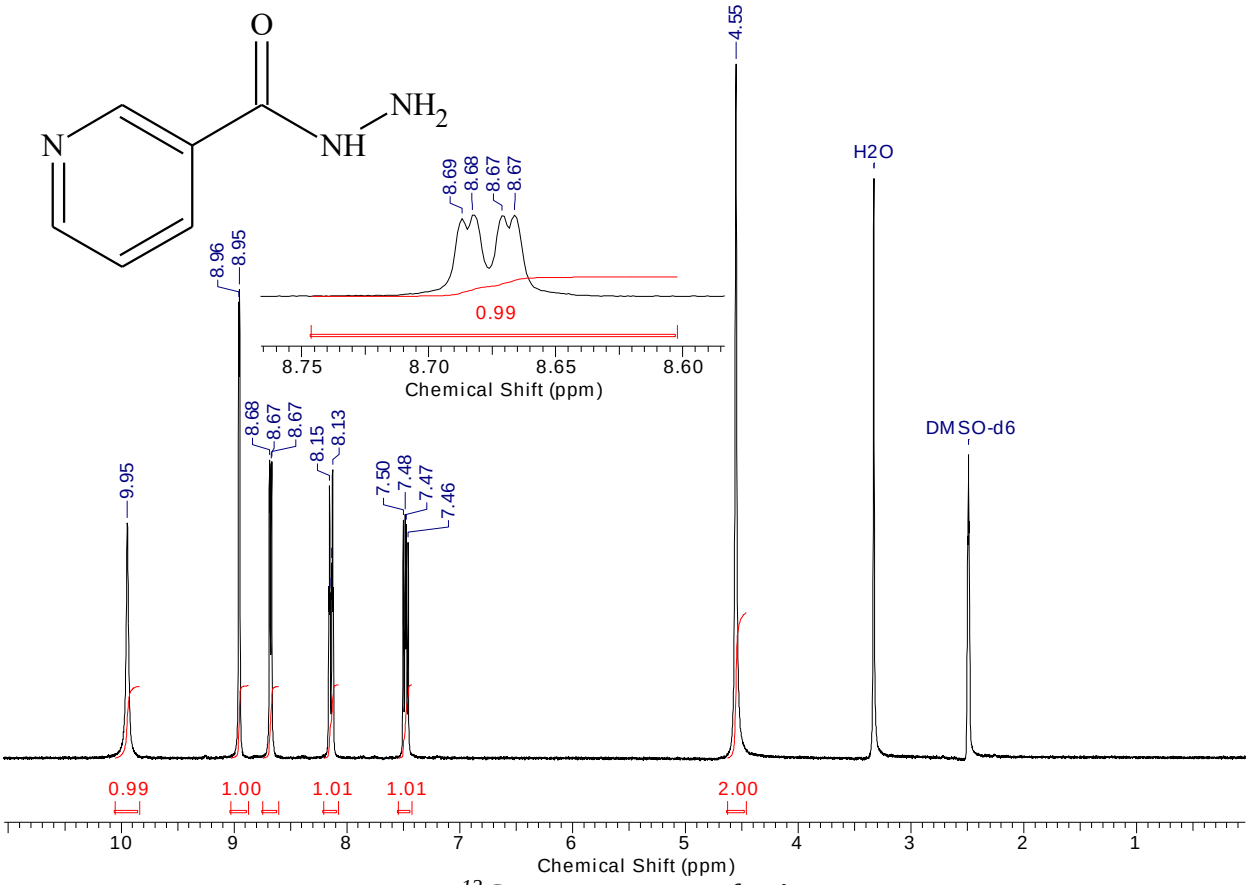
*^{1}H NMR spectrum of **11i***



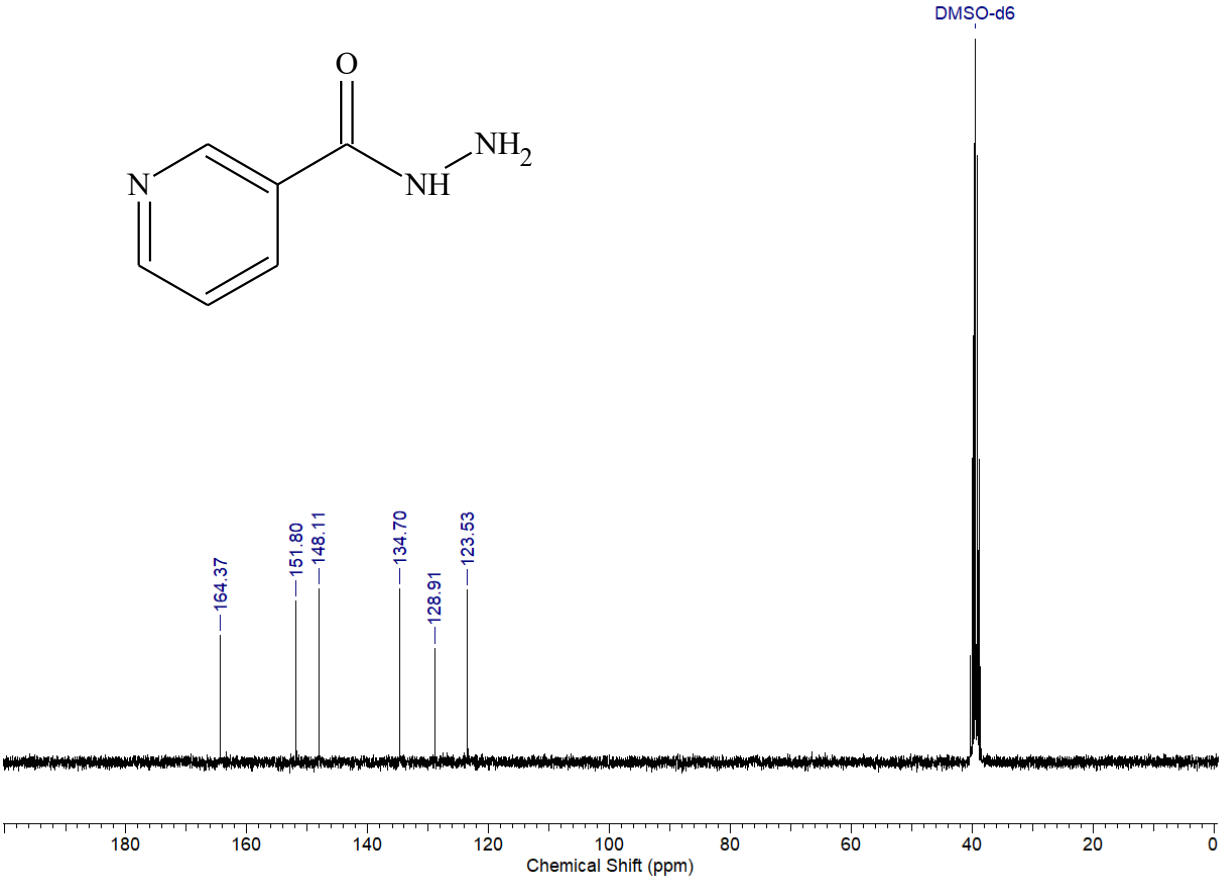
*^{13}C NMR spectrum of **11i***



*^{1}H NMR spectrum of **11j***



*^{13}C NMR spectrum of **11j***



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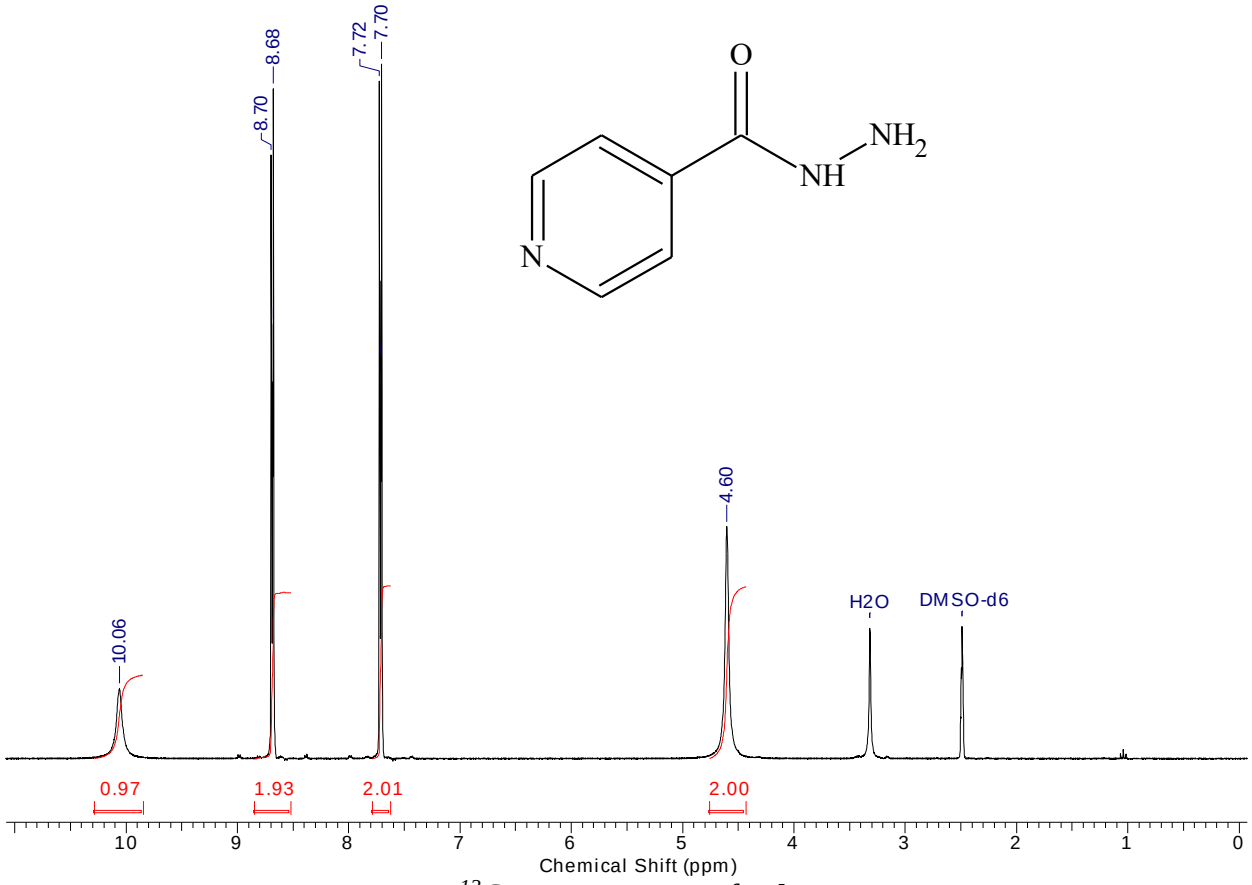

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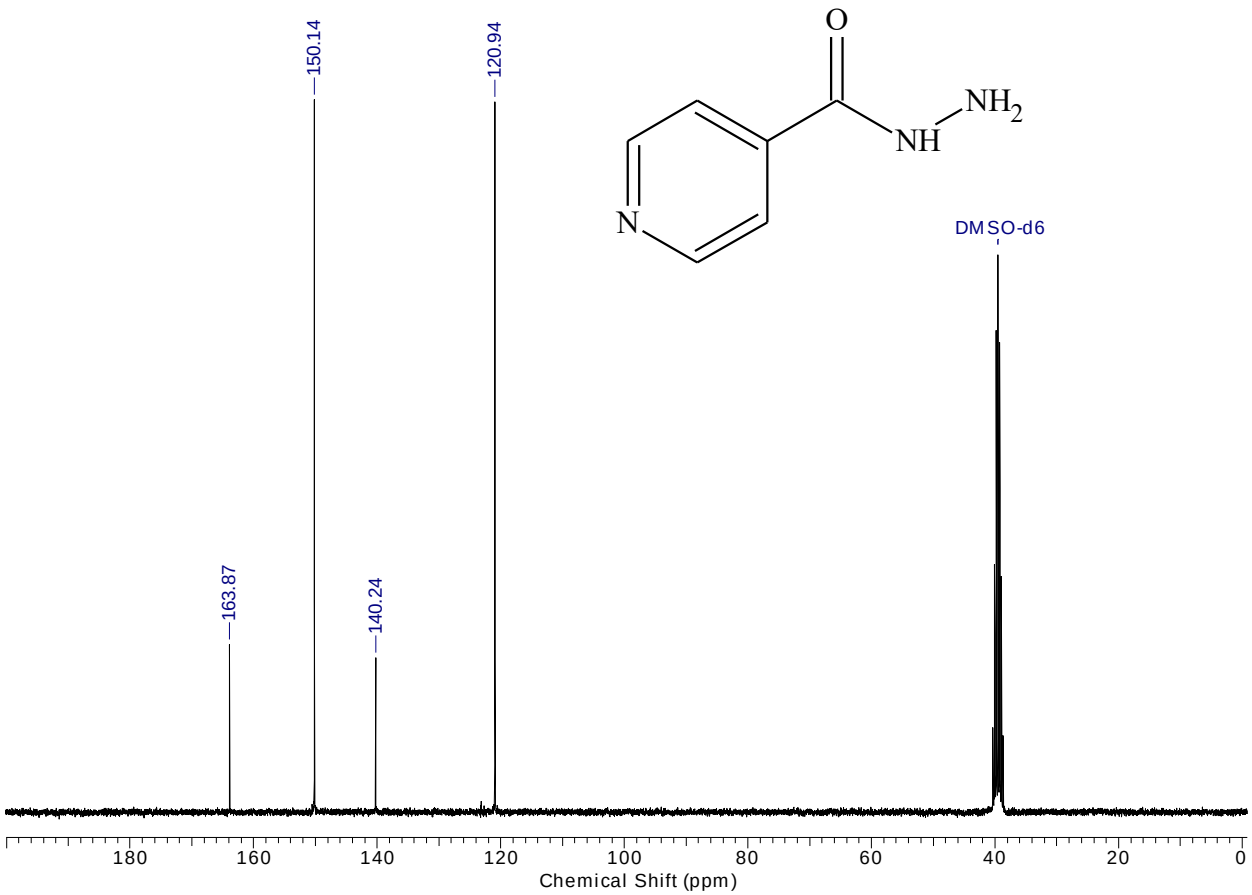
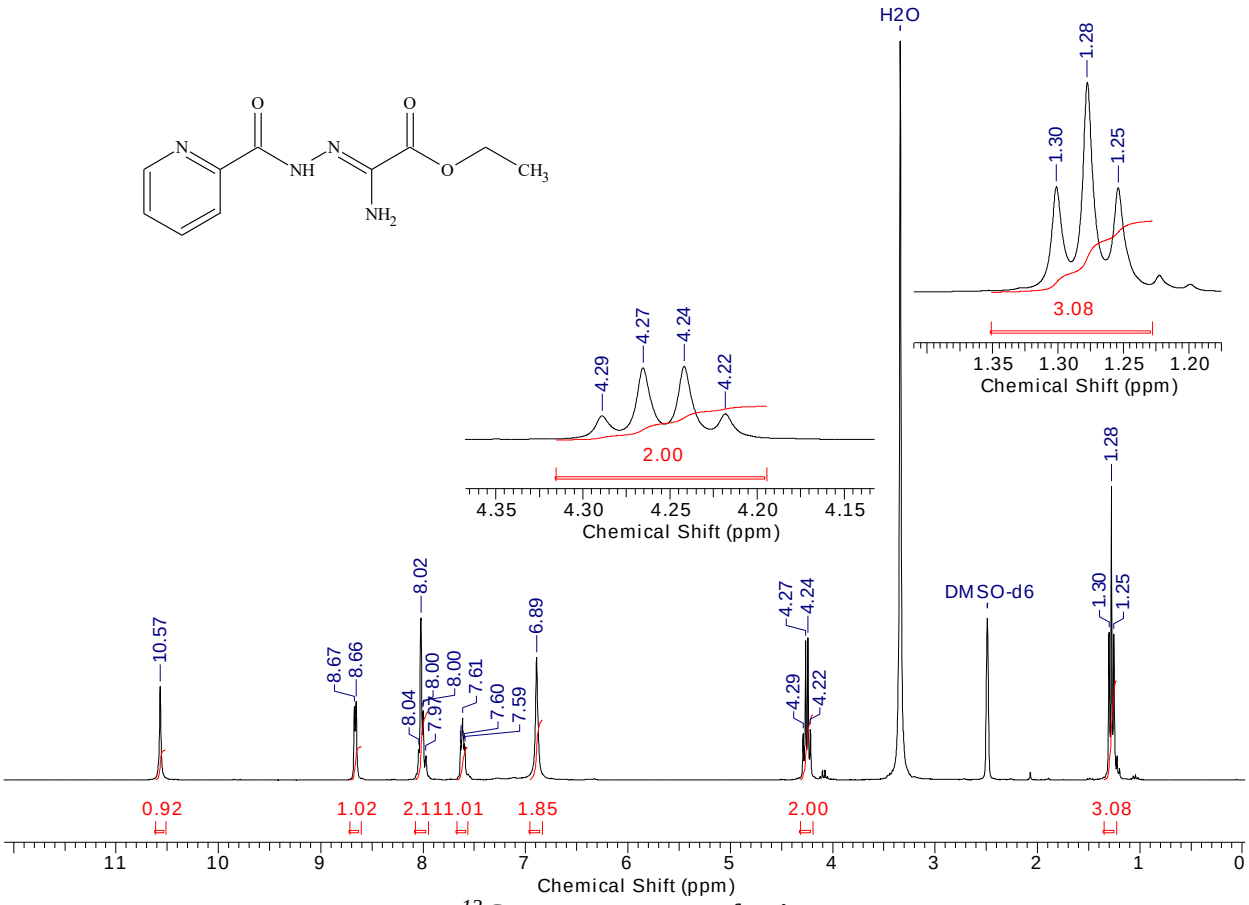
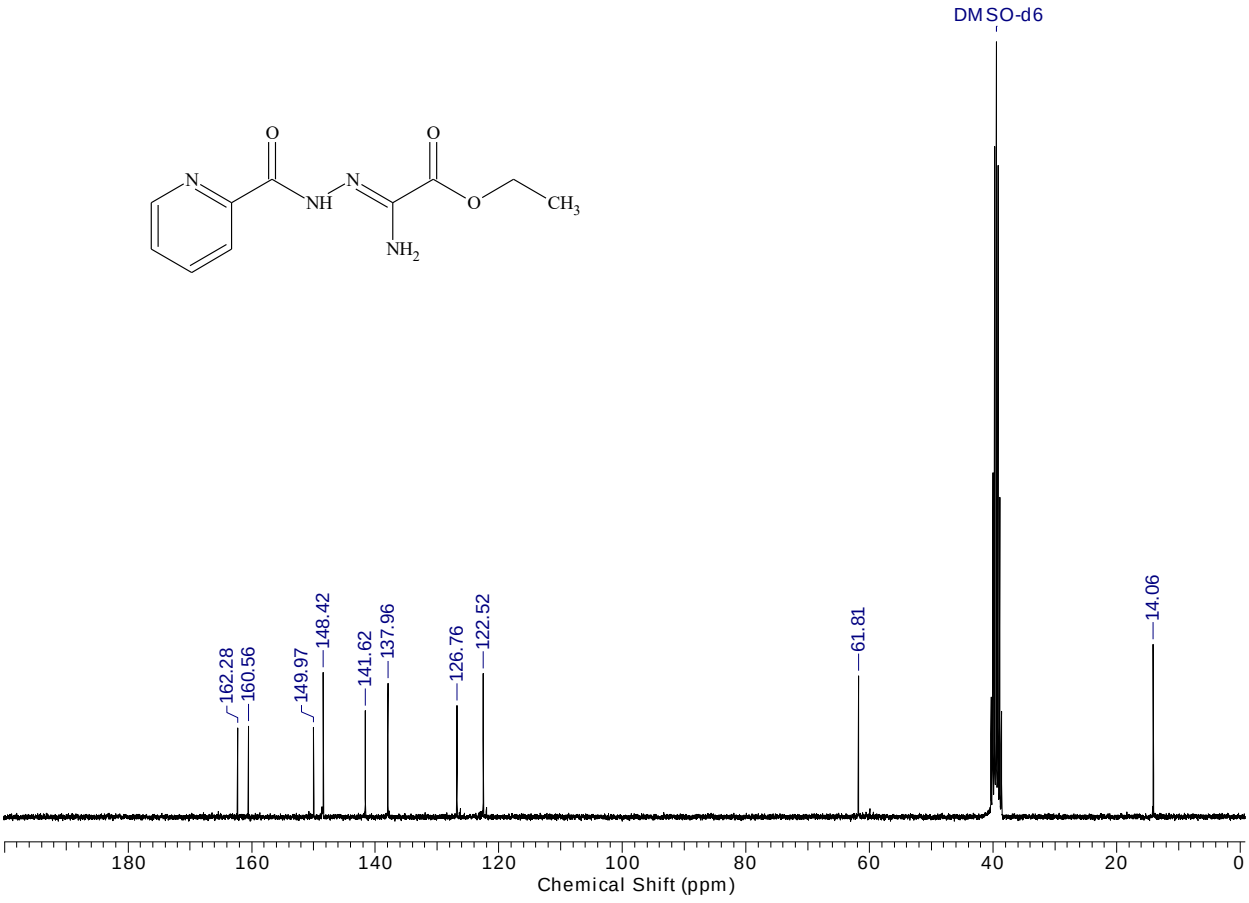


^{1}H NMR spectrum of ***12i***



^{13}C NMR spectrum of ***12i***



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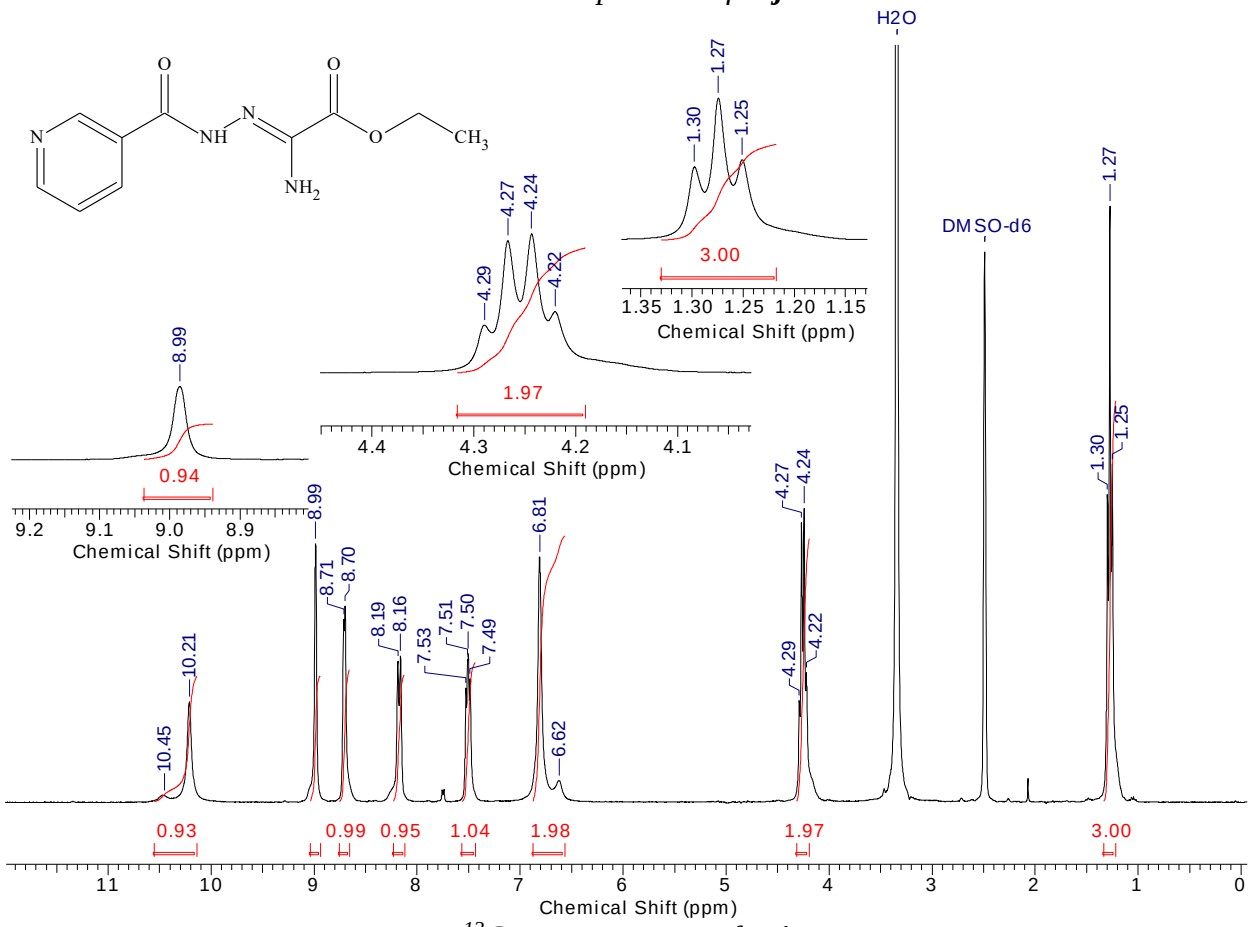
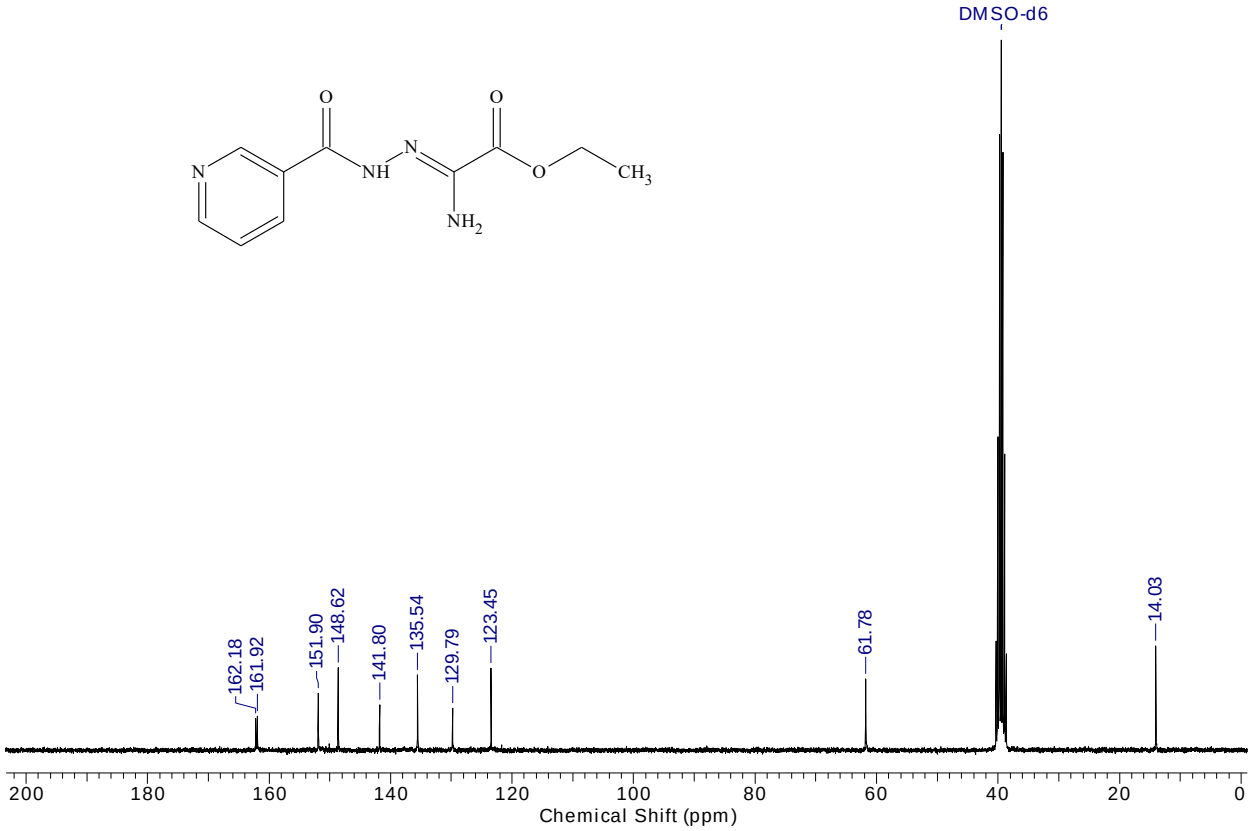


*^{13}C NMR spectrum of **12j***



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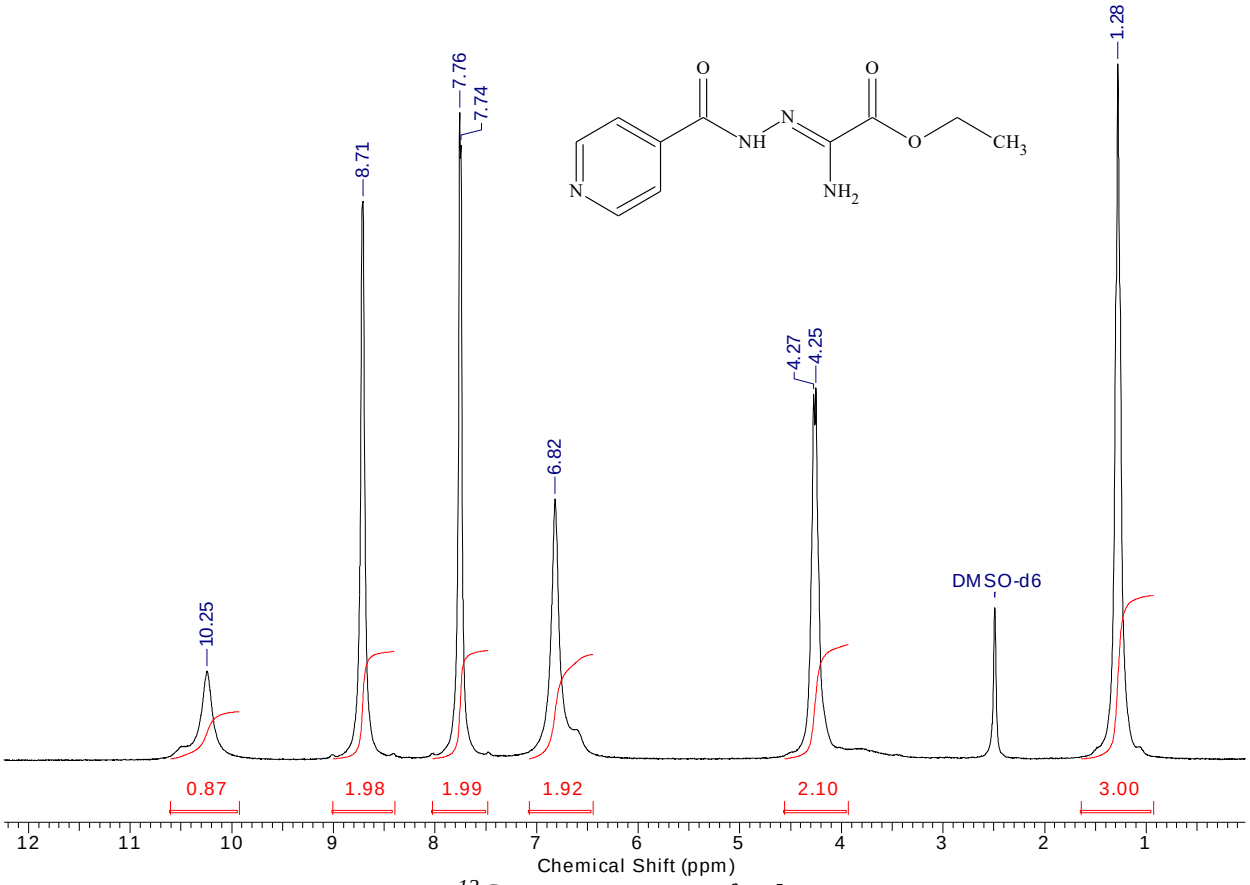

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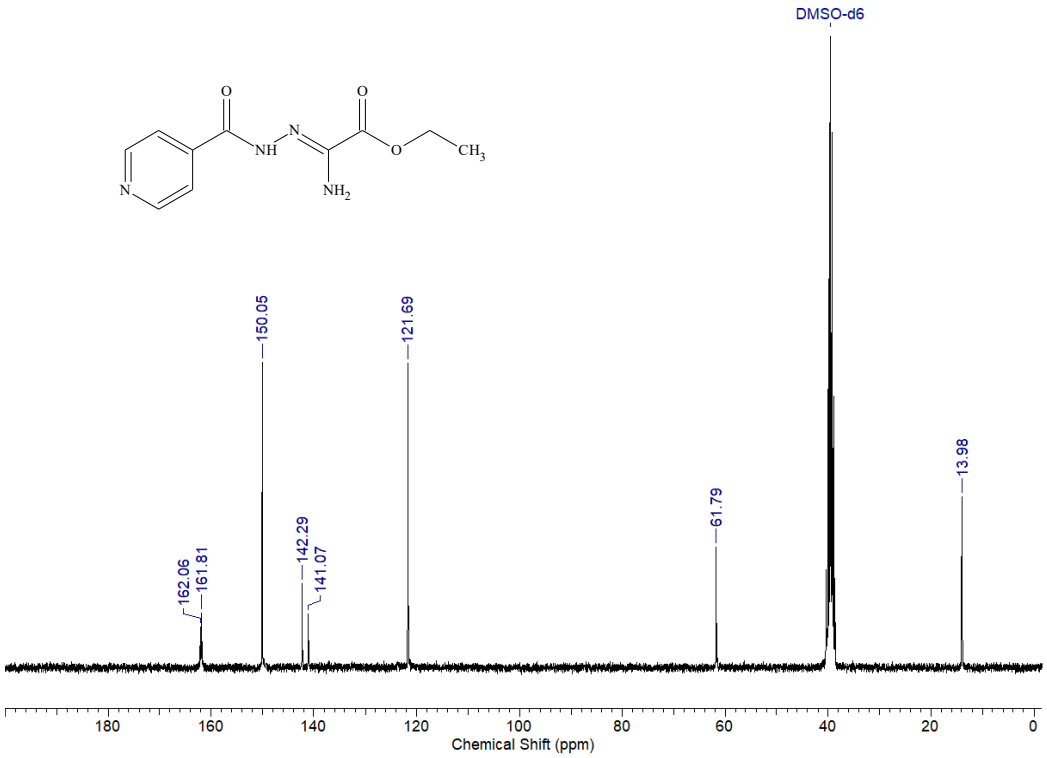
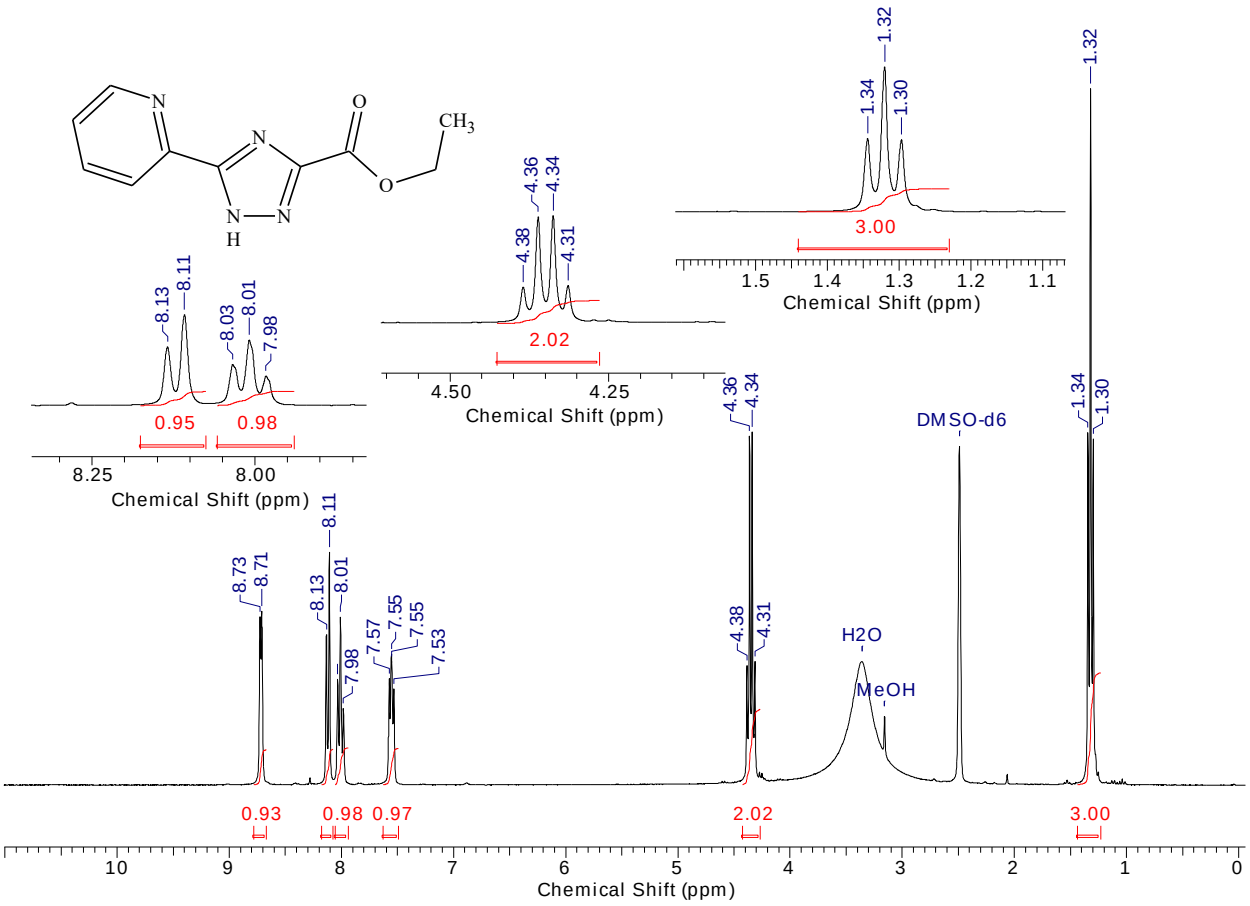
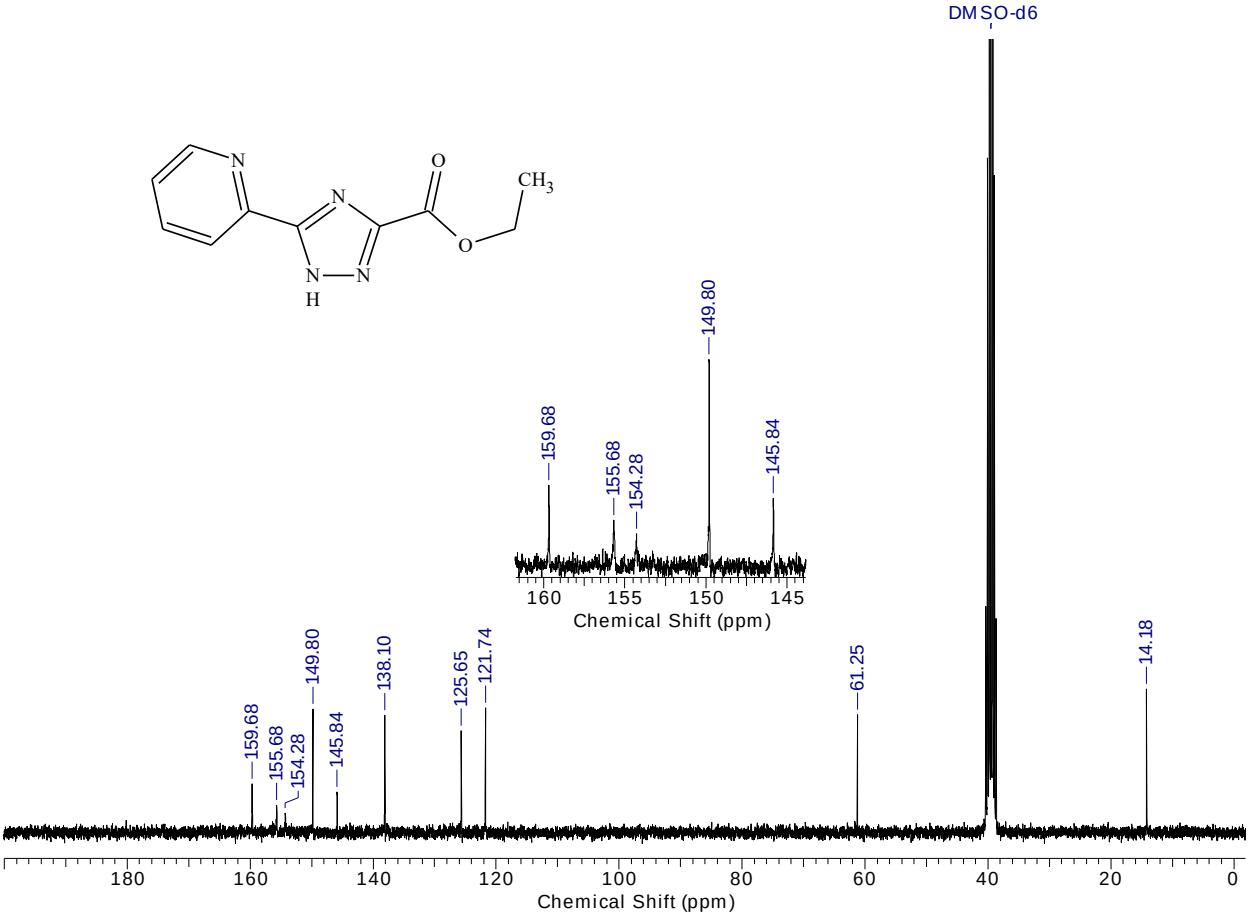


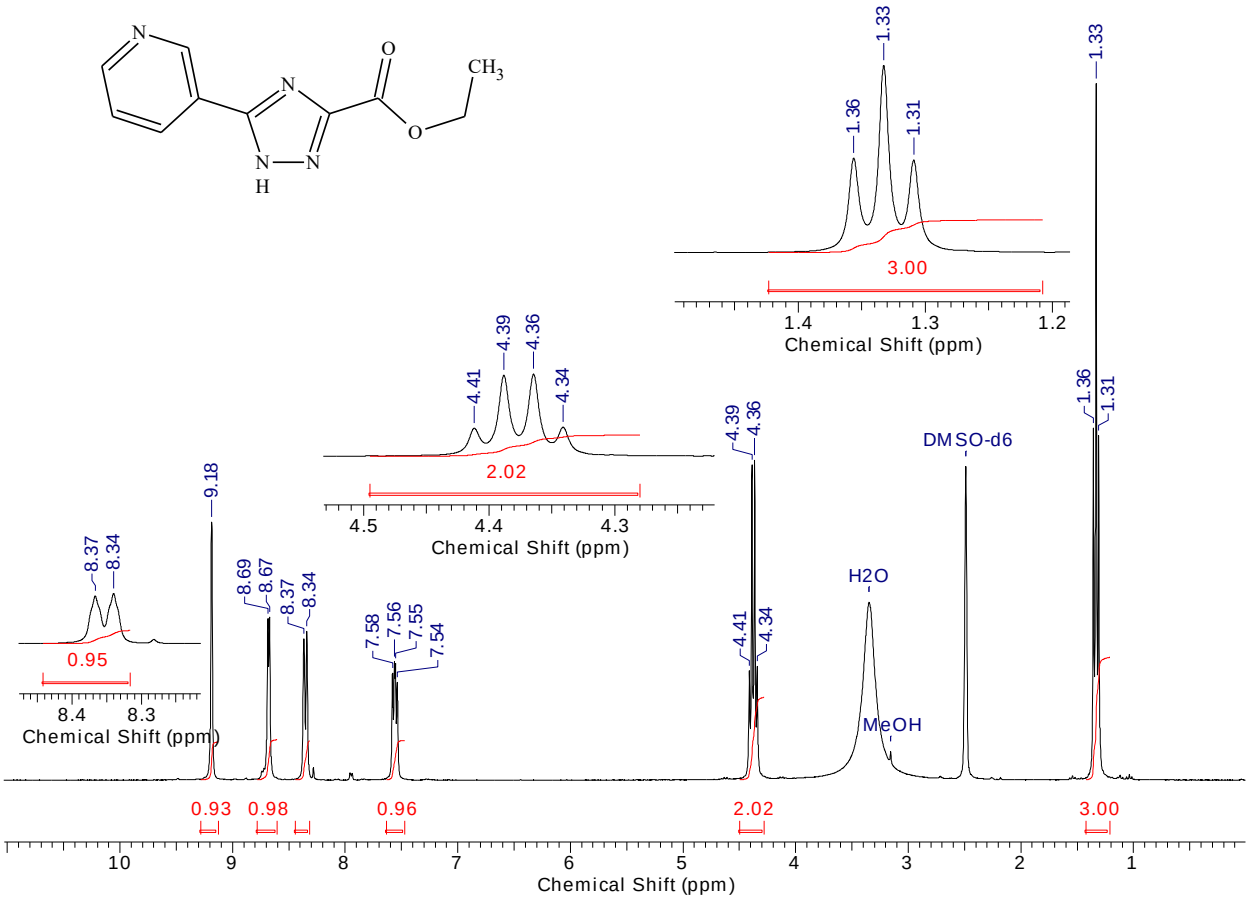
*^{1}H NMR spectrum of **13i***



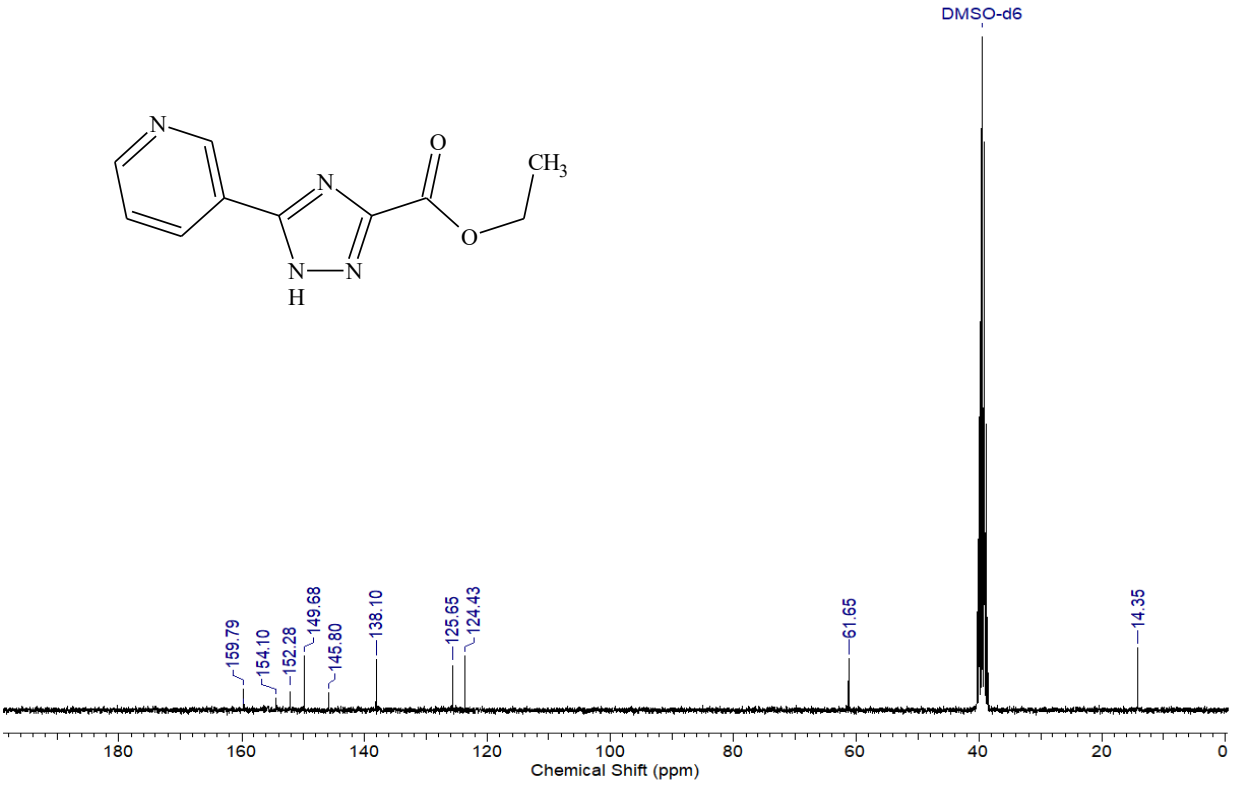
*^{13}C NMR spectrum of **13i***



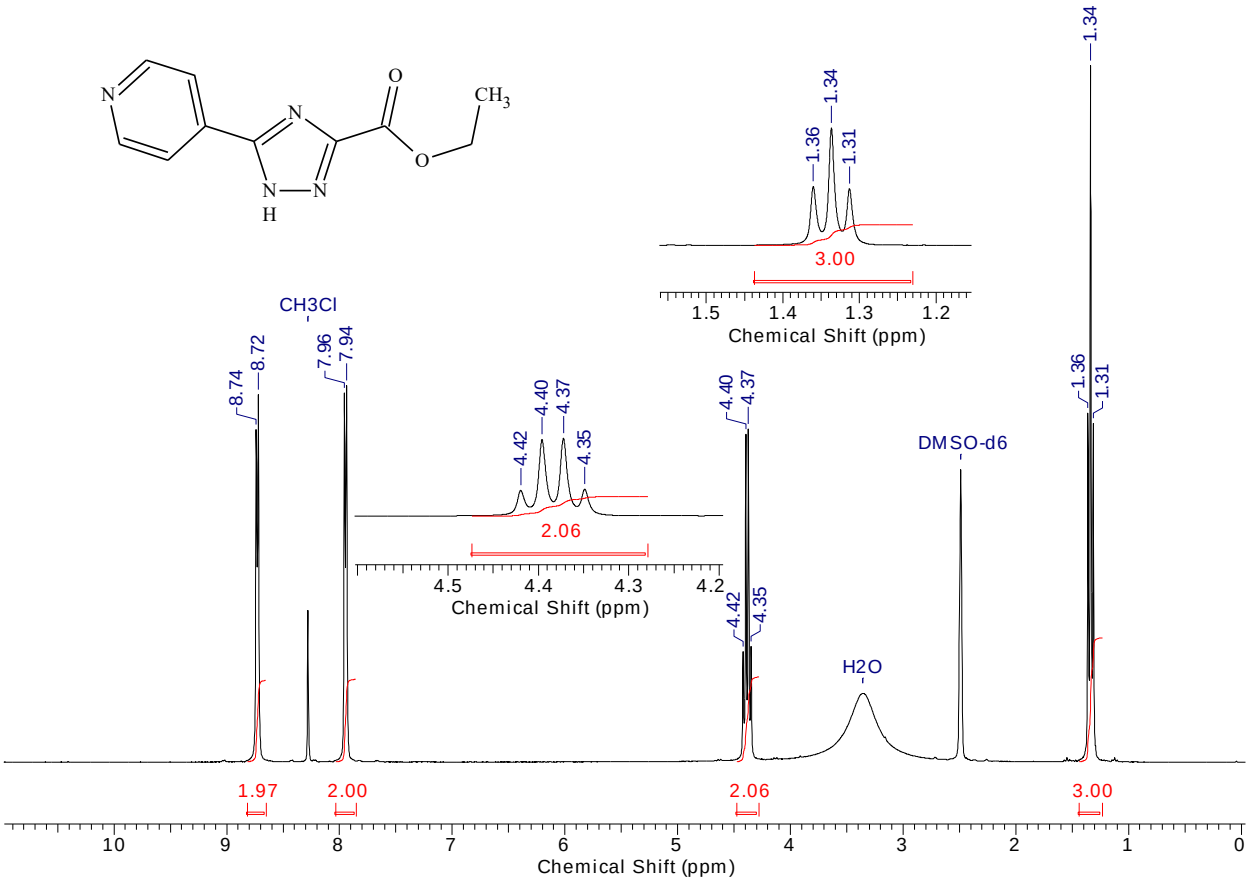
*^{1}H NMR spectrum of **13j***



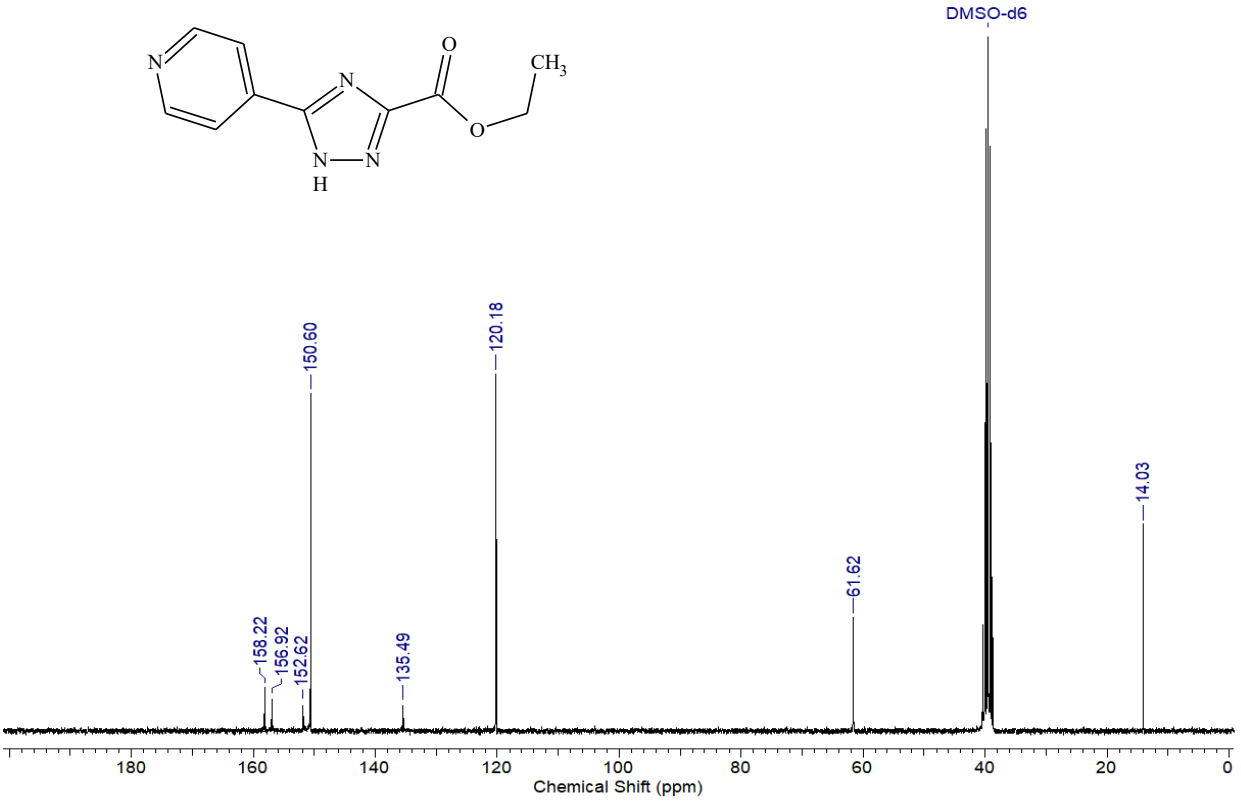
*^{13}C NMR spectrum of **13j***



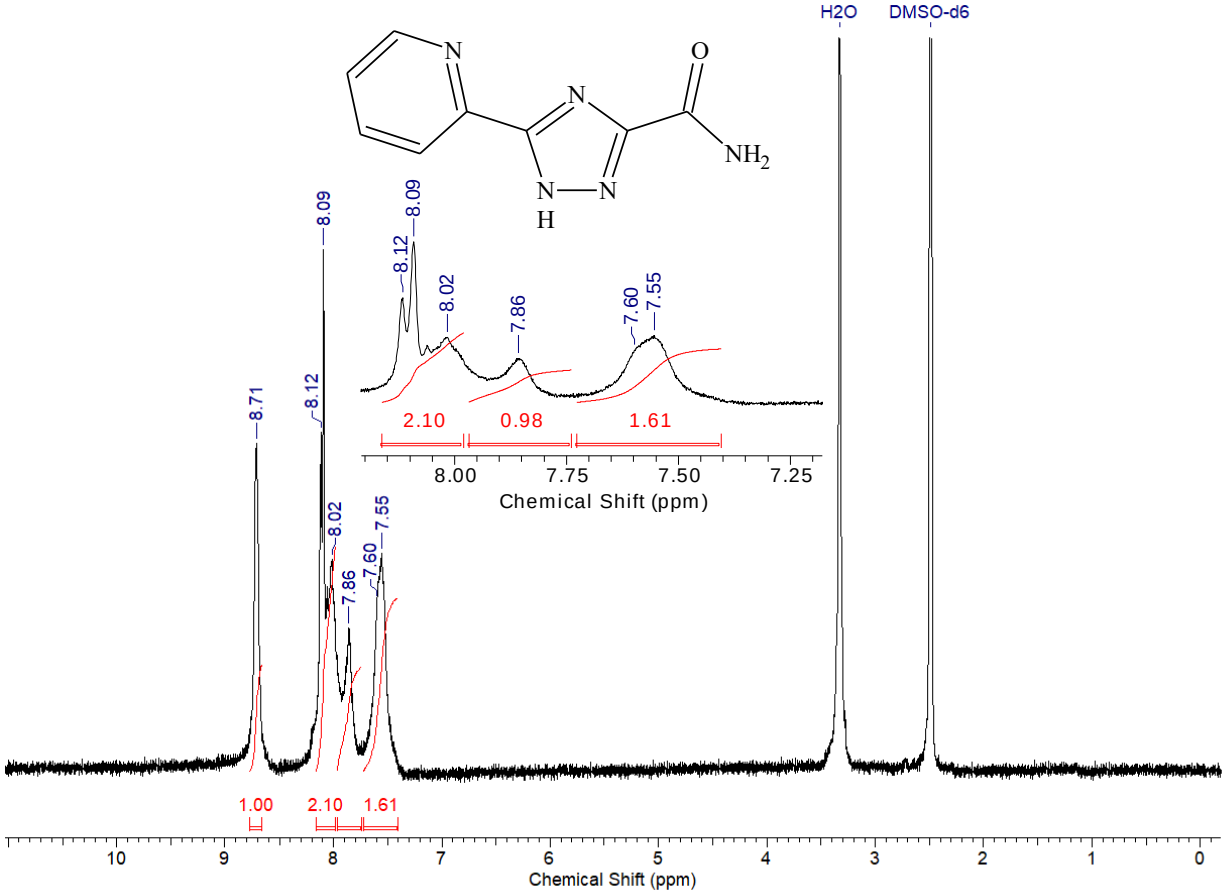
*^{1}H NMR spectrum of **13k***



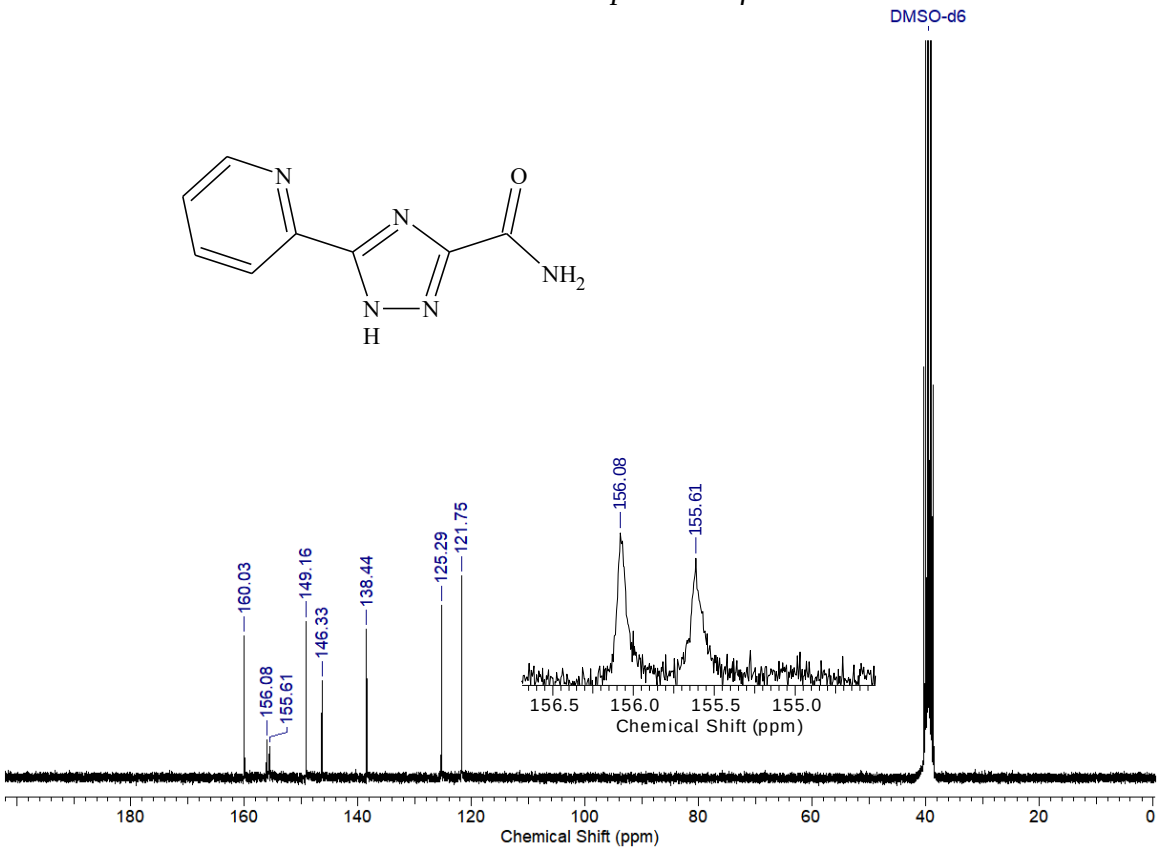
*^{13}C NMR spectrum of **13k***



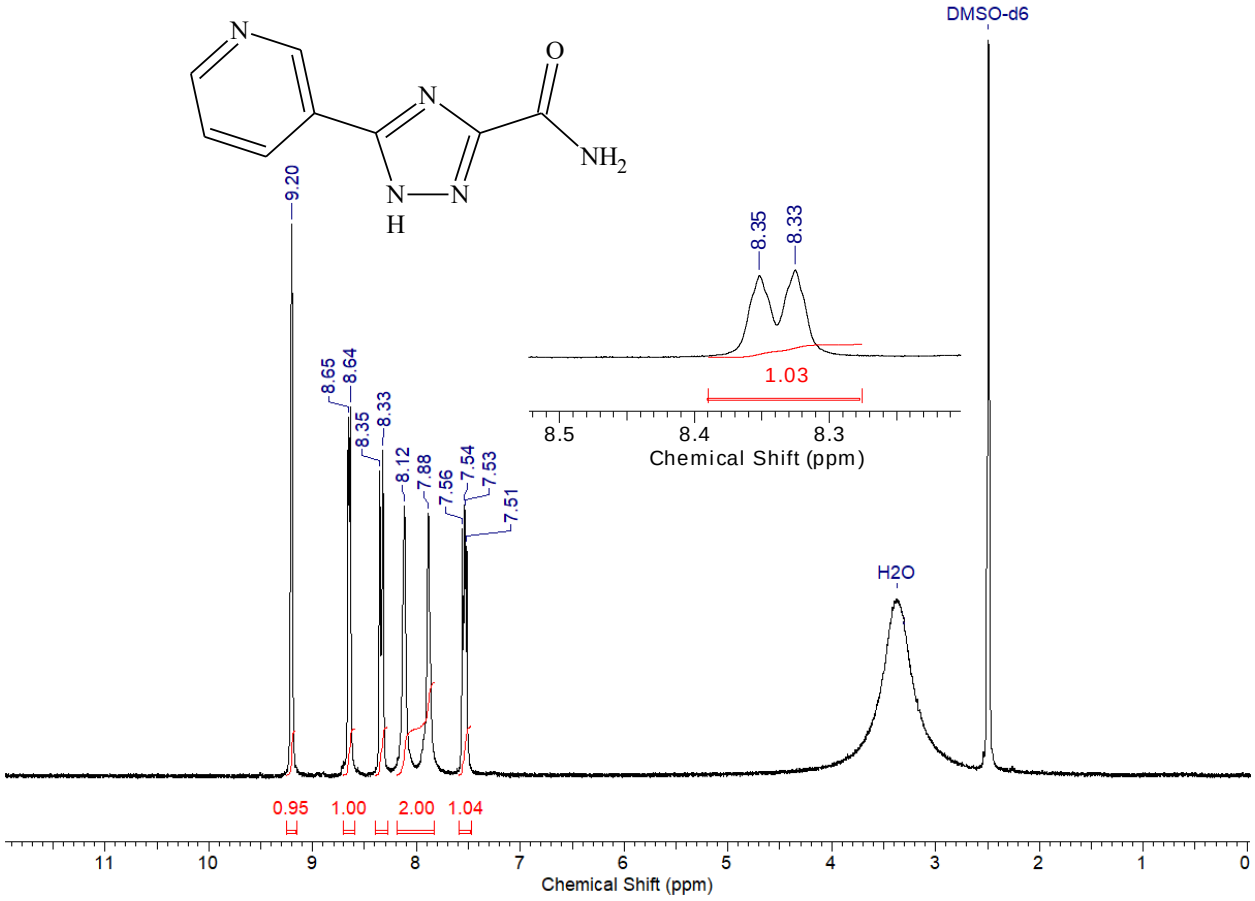
^{1}H NMR spectrum of ***4i***



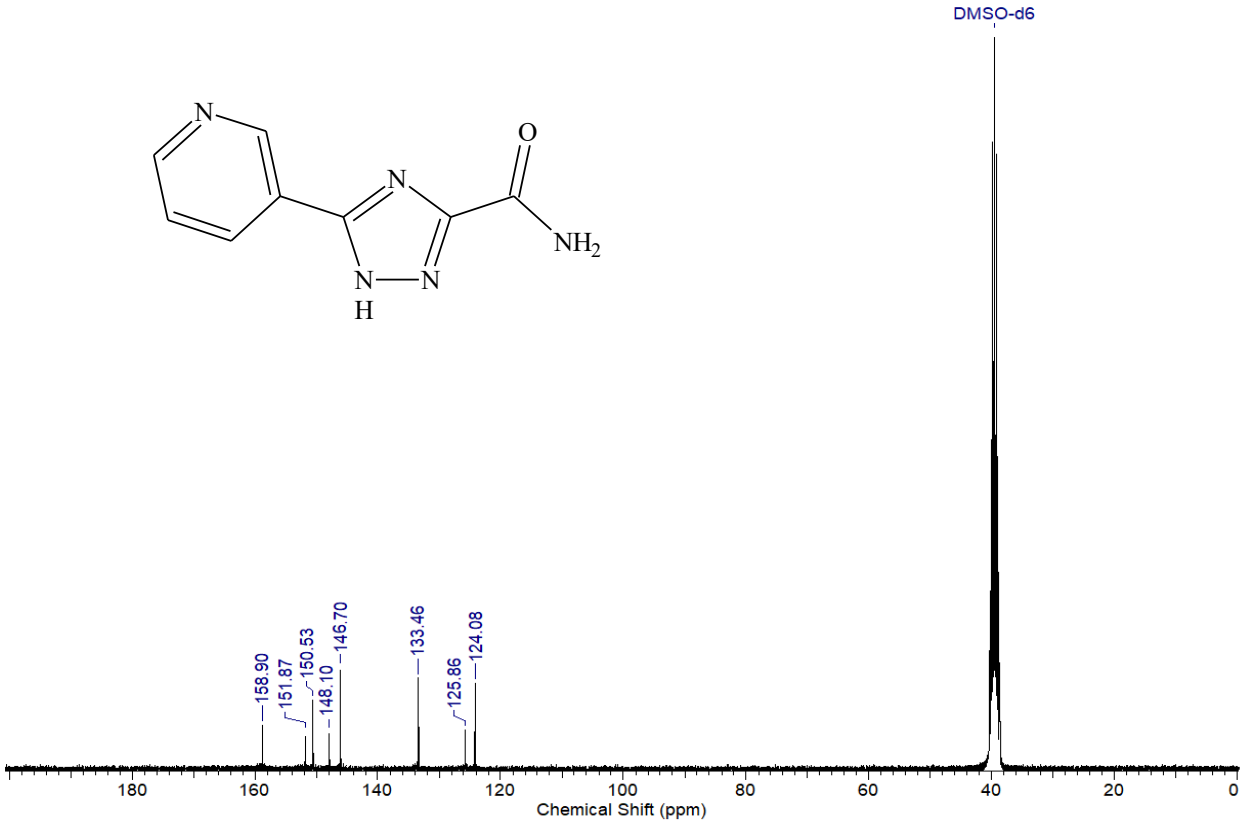
^{13}C NMR spectrum of ***4i***



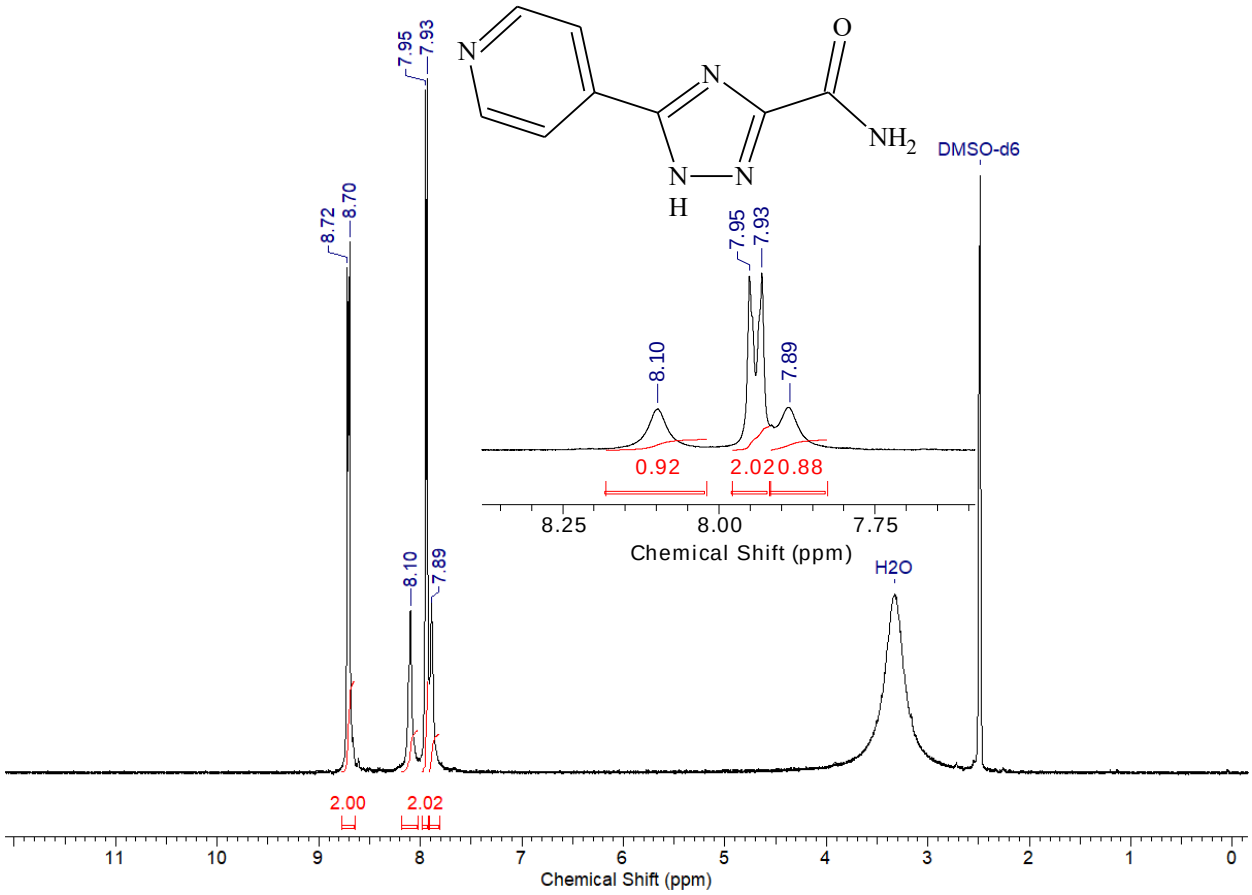
*^{1}H NMR spectrum of **4j***



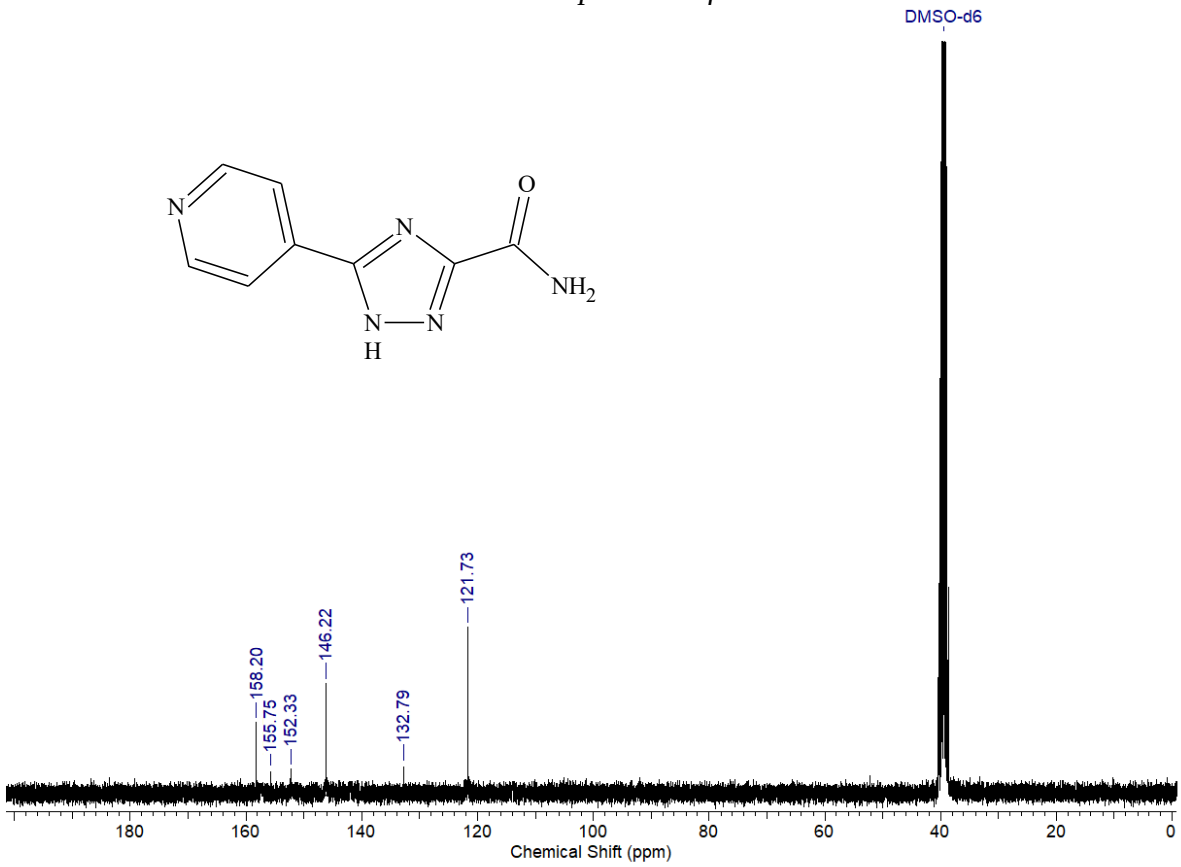
*^{13}C NMR spectrum of **4j***



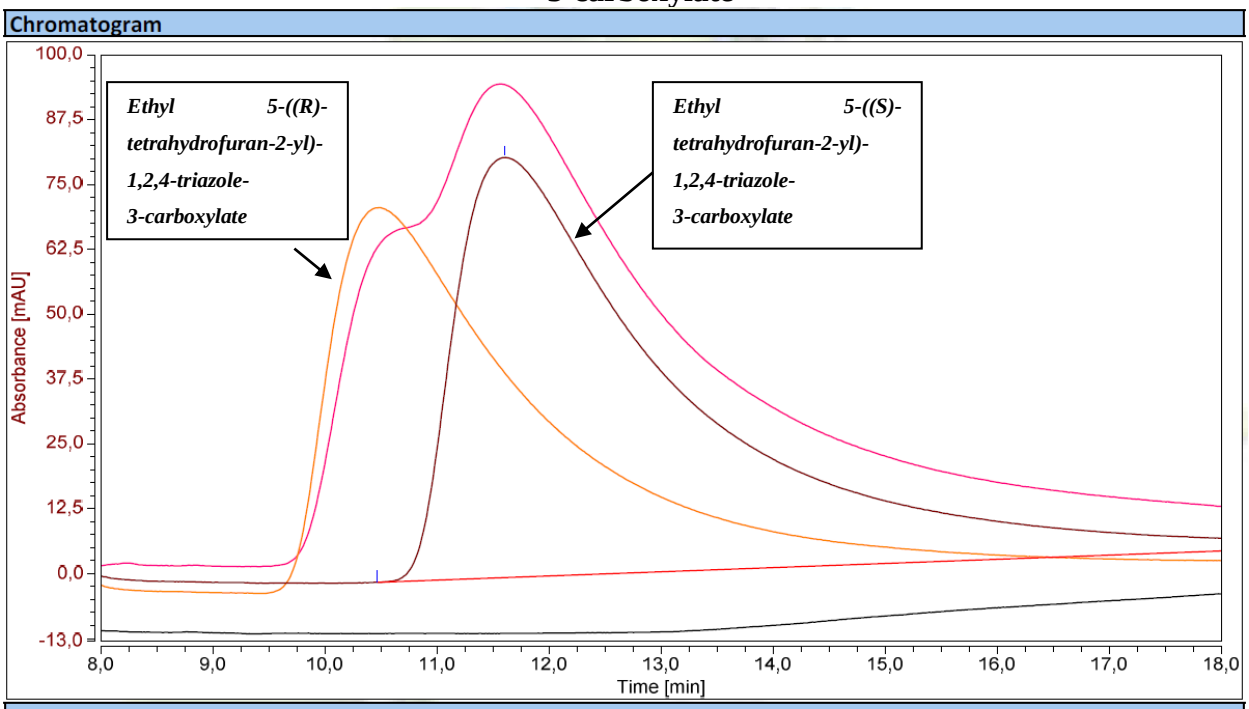
^{1}H NMR spectrum of ***4k***



^{13}C NMR spectrum of ***4k***



Chiral column chromatography of **ethyl 5-((*R*)-tetrahydrofuran-2-yl)-1,2,4-triazole-3-carboxylate** and **ethyl 5-((*S*)-tetrahydrofuran-2-yl)-1,2,4-triazole-3-carboxylate**



Integration Results

No.	Peak Name	Retention Time min	Area mAU*min	Height mAU	Relative Area %	Relative Height %	Amount n.a.
1		11,606	191,865	80,948	100,00	100,00	n.a.
Total:			**191,865**	**80,948**	**100,00**	**100,00**	

The chiral column chromatography of compounds ethyl 5-((*R*)-tetrahydrofuran-2-yl)-1,2,4-triazole-3-carboxylate and ethyl 5-((*S*)-tetrahydrofuran-2-yl)-1,2,4-triazole-3-carboxylate was carried out on HPLC system "Dionex UltiMate 3000" (Termo Scientific), consisting of a pump, a degasser, an autosampler, a thermostat and a didode array detector (DAD) on a 4.6 × 250 mm column filled with silica gel modified with 4-methyl benzoate, with a particle size of 10 µm (Chiralcel OJ (Chiral Technologies Inc.)). Column temperature (30 ± 1)°C, detector wavelength 210 nm, the volume of the injected sample is 50 µl, mobile phase 2-propanol : n-hexane (1:9), flow rate 0.5 ml/min.

LCMS purity of antiviral tested compounds

Table S1. *LCMS data*

Compound	Purity by HPLC, %	MS
2a	98	for $C_7H_{10}N_4O_2$ $[M+H]^+$ calculated: 183.18, found: 183.20; $[M-H]^-$ calculated: 181.17, found: 181.20.
2b	97	for $C_7H_{10}N_4O_2$ $[M+H]^+$ calculated: 183.18, found: 183.20; $[M-H]^-$ calculated: 181.17, found: 181.19.
3a	98	for $C_7H_{10}N_4O_2$ $[M+H]^+$ calculated: 183.18, found: 183.20.
3b	96	for $C_8H_{12}N_4O_2$ $[M+H]^+$ calculated: 197.21, found: 197.22.
4a	96	for $C_7H_{10}N_4O_2$ $[M+H]^+$ calculated: 183.18, found: 183.19; $[M-H]^-$ calculated: 181.17, found: 181.19.
4b	97	for $C_8H_{12}N_4O_2$ $[M+H]^+$ calculated: 197.21, found: 197.22; $[M-H]^-$ calculated: 195.19, found: 195.20.
4c	97	for $C_8H_{12}N_4O_2$ $[M+H]^+$ calculated: 197.21, found: 197.22; $[M-H]^-$ calculated: 195.19, found: 195.20.
4d	95	for $C_8H_{12}N_4O_2$ $[M+H]^+$ calculated: 197.21, found: 197.23; $[M-H]^-$ calculated: 195.19, found: 195.21.
4e	96	for $C_7H_6N_4O_2$ $[M+H]^+$ calculated: 179.15, found: 179.16; $[M-H]^-$ calculated: 177.14, found: 177.16.
4f	98	for $C_7H_6N_4O_2$ $[M+H]^+$ calculated: 179.15, found: 179.16; $[M-H]^-$ calculated: 177.14, found: 177.16.
4g	96	for $C_7H_6N_4OS$ $[M+H]^+$ calculated: 195.22, found: 195.24; $[M-H]^-$ calculated: 193.20, found: 193.22.
4h	98	for $C_7H_6N_4OS$ $[M+H]^+$ calculated: 195.22, found: 195.23; $[M-H]^-$ calculated: 193.20, found: 193.21.
4i	98	for $C_8H_7N_5O$ $[M+H]^+$ calculated: 190.18, found: 190.19; $[M-H]^-$ calculated: 188.17, found: 188.17.
4j	97	for $C_8H_7N_5O$ $[M+H]^+$ calculated: 190.18, found: 190.20; $[M-H]^-$ calculated: 188.17, found: 188.19.
4k	97	for $C_8H_7N_5O$ $[M+H]^+$ calculated: 190.18, found: 190.19; $[M-H]^-$ calculated: 188.17, found: 188.18.
5	96	for $C_8H_{12}N_4O_2$ $[M+H]^+$ calculated: 197.21, found: 197.23; $[M-H]^-$ calculated: 195.20, found: 195.21.
6	96	for $C_9H_{14}N_4O_3$ $[M+H]^+$ calculated: 227.24, found: 227.25; $[M-H]^-$ calculated: 225.22, found: 225.23.
7	97	for $C_{11}H_{16}N_4O_2$ $[M+H]^+$ calculated: 237.28, found: 237.30; $[M-H]^-$ calculated: 235.26, found: 235.27.
8	96	for $C_7H_{10}N_4OS$ $[M+H]^+$ calculated: 199.25, found: 199.28; $[M-H]^-$ calculated: 197.24, found: 197.27.

In vitro studies

Cytopathic effect (CPE) inhibition assay against SARS-CoV2

Vero E6 cells (ATCC CRL-1586) were seeded in 96-well plates at a density of 2×10^4 cells per well in complete DMEM medium (Gibco, Thermo Fisher Scientifc, Waltham, USA), supplemented with 10% fetal bovine serum (FBS; HyClone, Cytiva, Marlborough, MA, USA), 1 × GlutaMAX-l (Gibco, Thermo Fisher Scientifc, Waltham, USA) and 1 × penicillin–streptomycin (PanEco, Russia). After 24 h of incubation (37°C, 5% CO_2) solutions of test compounds in growth medium were added to the cell monolayer to a 500 μM concentration in the wells. After 1 h of incubation, 100 $TCID_{50}$ SARS-CoV2 (PMVL-4, GISAID EPI_ISL_470898) was added to the cell monolayer in the absence or presence of test compounds. After 72 h of incubation, differences in cell viability caused by virus-induced cytopathic effects were analyzed using the MTT method as previously described.[S3,S4] For this, an MTT (Sigma, Merck KGaA, Darmstadt, Germany) stock solution (5 mg/ml in PBS) was added to each well at a final concentration of 0.5 mg/ml. After a 2 h incubation, medium was aspirated from wells and 150 μl of DMSO was added. Absorbance was measured at 590 nm using a SPECTROstar Nano microplate reader (BMG LABTECH GmbH, Ortenberg, Germany). Three independent experiments with triplicate measurements were performed.

Pseudovirus assay

In this assay with pseudo-SARS-CoV2 (lineage B.1.1.7),[S5] 293T/ACE2 cells were used. The test compounds in a concentration in the well of 500 μM were added to the cells except for control. After 1 h of incubation, cells were transduced with pseudo-SARS-CoV2. After incubation for 72 h, the fluorescence of GFP was determined using a microplate reader (Ex 488nm / Em 510nm) (Hidex Sense Beta Plus, Hidex, Turku, Finland).

Table S2. *In vitro* tests results

Compound	% inhibition of SARS-CoV2 CPE (in concentrations of 0.5 mM)	% inhibition on the model of pseudo-SARS-CoV2 (in concentrations of 0.5 mM)
1	41 ± 4	-
2a	-	-
2b	49 ± 6	35 ± 7
3a	16 ± 6	-
3b	57 ± 5	-
4a	24 ± 8	15 ± 5
4b	31 ± 1	-
4c	-	-
4d	-	-
4e	14 ± 6	-
4f	-	-
4g	-	-
4h	-	-
4i	24 ± 6	-
4j	25 ± 5	-
4k	40 ± 7	100 - 9
5	22 ± 5	-
6	36 ± 6	-
7	21 ± 4	-
8	65 ± 8	100 - 14

In silico studies

1. Ligands 3D structure preparation.

The geometries of compounds were minimized by energy using an MM2 force field in Chem3D software (Perkin Elmer Informatics, Inc., Waltham, MA, USA).

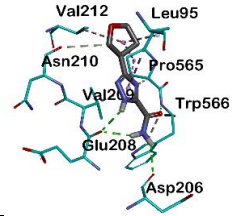
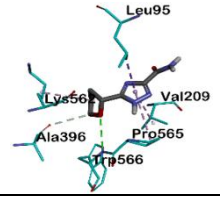
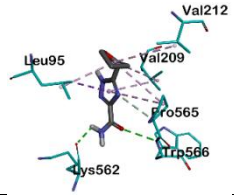
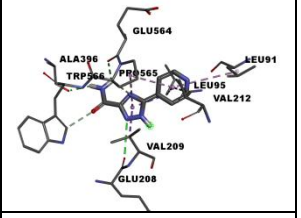
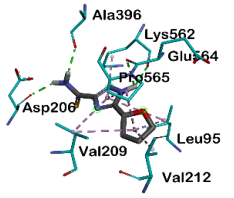
2. Protein 3D structure preparation.

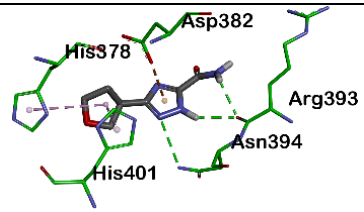
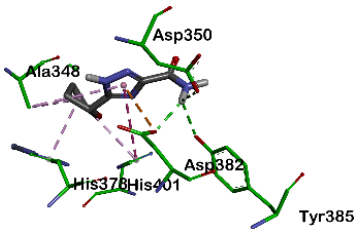
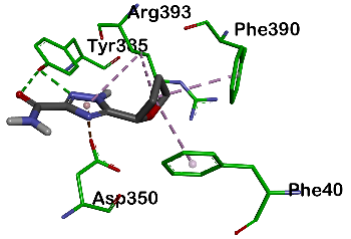
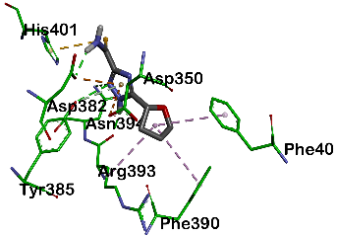
The carboxypeptidase ACE2 (EC no. 3.4.17.23) crystal structure was obtained from the Protein Data Bank (PDB ID: 1R4L). The selected structure of the complex has a resolution of 3.0 Å and does not contain breaks in the main chain of the protein near the orthosteric or allosteric centers. Hydrogen atoms addition, atom types assignment, nonpolar hydrogen atoms unification, partial Gasteiger and Kollmann charges calculation were carried out using AutoDockTools 1.5.7 (The Scripps Research Institute, La Jolla, CA, США). Solvent molecules were removed using UCSF Chimera 1.15.

3. Molecular Docking Calculations

All of the torsional bonds of the ligands were free to rotate, while protein was held rigid. A grid box of dimensions 40 × 40 × 40 with a spacing of 1 Å was created and centered on the orthosteric site. The Autodock 4.2.6 (The Scripps Research Institute, La Jolla, CA, USA) was used for docking calculations involve the Lamarckian genetic algorithm (LGA).[S6] Ligand interactions were determined, and graphical representations were made using the Discovery Studio Visualizer (version 21.1.0.20298, Dassault Systems Biovia Corp., San Diego, CA, USA).

Table S3. Interactions of compounds **3a**, **3b**, **4a**, 4**k** and **8** with allosteric binding sites 2 and 3 AR.

Compound	Affinity, kcal/mol	Aminoacids of the allosteric site 2 (AS_2)																				Aminoacids out of a site	Interactions
4a	-6,5	396	397	392	563	564	562	566	565	391	206	212	209	208	91	96	95	210	99	98	94	97	88	85		
2a	-5,5	396	397	392	563	564	562	566	565	391	206	212	209	208	91	96	95	210	99	98	94	97	88	85		
2b	-5,8	396	397	392	563	564	562	566	565	391	206	212	209	208	91	96	95	210	99	98	94	97	88	85		
4k	-6.4	396	397	392	563	564	562	566	565	391	206	212	209	208	91	96	95	210	99	98	94	97	88	85		
8	-6,1	396	397	392	563	564	562	566	565	391	206	212	209	208	91	96	95	210	99	98	94	97	88	85	207	

Compound	Affinity, kcal/mol	Aminoacids of the allosteric site 3 (AS_3)													Aminoacids out of a site					Interactions
4a	-5,8	356	355	382	354	350	351	385	386	352	353	393	390	40	37	394	401	378			
2a	-5,9	356	355	382	354	350	351	385	386	352	353	393	390	40	37	394	401	378	348	349	
2b	-6	356	355	382	354	350	351	385	386	352	353	393	390	40	37	394	401				
8	-5,8	356	355	382	354	350	351	385	386	352	353	393	390	40	37	394	401				

Legend: types of possible interactions[S7]							
	Classic H		Classic H + hydrophobic		Other H		Ionic		Hydrophobic
	2 classic H		Classic H + ionic		Other H + hydrophobic		Van der Waals		

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