Supplementary Information

Rapid Assembly of Potent Type II Dehydroquinase Inhibitors *via* "Click" Chemistry

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Molecular modeling experimental

Protein Preparation

The crystal structures of *S. coelicolor* and *H. pylori* type II dehydroquinase were downloaded from the Research Collaboratory for Structural Bioinformatics (RCSB) Protein DataBank (PDB code: 1GU1 and 2WKS)[1, 2] respectively. The structures were subjected to further modification to ensure suitability for molecular docking using Glide (version 5.0).[3] The ligand/protein co-crystal structures were imported into Accelrys DS visualiser 2.0 (Accelrys Software Inc.) where the dodecamers were simplified into a dimer (Subunits A and B). Glycerol and 2,3-anhydroquinic acid for 1GU1 and CB6 for 2WKS were kept in sub-unit A . Structural waters were also removed except for H_2O-2227 in sub-unit A for 1GU1. The simplified type II dehydroquinase protein structures were then imported into Maestro (version 9.0) and prepared using the *Protein Preparation Wizard* tool in which bond orders were assigned to the ligands. Hydrogens were added to both ligands and the protein in a manner consistent with physiological pH (pH = 7.0) using an all atom force field. Restrained minimization of the protein structure was then conducted using *impref* using an OPLS-AA force field until the root mean square deviation (RMSD) of non-hydrogen atoms reached 0.3.

Inhibitor Preparation

Inhibitors were built in Maestro (version 9.0) and used as maegz files. Hydrogens were added to each structure. Inhibitors were then subjected to the *geometry cleanup* utility which minimises the energy of structures using a Universal Force Field (UFF).

Glide docking

A receptor grid file was generated using the *Receptor Grid Generation* utility in Glide (version 5.0). The ligands chosen were 2,3-anhydroquinic acid for 1GU1 and CB6 for 2WKS as the centre of receptor grid generation. Van der Waal radius scaling and partial charge cut off default values were used. Per atom scale factors which soften receptor potential were not used. After the receptor grids were generated, ligands were docked into the active site using extra precision mode (XP).

Biological Experimental

Purification of *S. coelicolor*, *H. pylori* and *M. tuberculosis* type II dehydroquinases

The *H. pylori* and *S. coelicolor aroQ* genes have been previously cloned into pET-15b vectors to facilitate purification and the *M. tuberculosis* aroD gene cloned into pET 28a was a gift from Prof. Chris Abell, University of Cambridge. Plasmids were transformed into BL21(DE3) competent cells and incubated overnight at 37° C on LB agar plates containing the appropriate concentration of the selection antibiotic (100μg/ml Ampicillin for pET-15b vectors and 30μg/ml kanamycin for the pET 28a vector). Single bacterial colonies were picked and inoculated into 500ml of autoinduction media[4] containing selection antibiotic and incubated at 37°C in the shaking incubator for a minimum of 24 hours. The cells were harvested and purified using Ni^{2+} affinity as described previously.[5]

Type II dehydroquinase assays

Enzyme assays for type II dehydroquinases from *S. coelicolor, H. pylori* and *M. tuberculosis* were carried out using a Shimadzu UV Vis1800 spectrometer with a 6×6 peltier cell holder using 1 cm path length quartz cuvettes at a wavelength of 234 nm to monitor the formation of the product, 3-dehydroshikimate. Specifically, the initial reaction rates were measured by the increase in absorbance at 234 nm, due to the formation of the enone-carboxylate chromophore of 3-dehydroshikimate ($\varepsilon = 1.2 \times 10^4$ M^{-1} cm^{-1}). The assays were performed at 25 ^{o}C in Tris-HCl buffer (0.05 M, pH 7.0) for *S. coelicolor* and *M. tuberculosis* or Tris-acetate (0.05 M, pH 7.0) for *H. pylori* type II dehydroquinases. The assays for *S. coelicolor*, *H. pylori* and *M. tuberculosis* type II dehydroquinases contained 0.62, 2.41 and 3.80 nM of the enzyme, and were performed in duplicate. The assay mixtures were prepared in 1 mL quartz cuvettes, and the assays were initiated by the addition of the substrate (3-dehydroquinate) to the mixture after incubating the buffer, inhibitor and enzyme at 25 ^{o}C for three minutes.

The kinetic data for inhibition studies were obtained by measuring the initial rates of reaction over a range of inhibitor concentrations (3-4 different concentrations) at 5-6 different substrate concentrations (0.1 K_M - 4 K_M). The inhibition constants (K_I) and the standard

deviations were determined using a non-linear regression fitting to the competitive model by GraphPad Prism (version 5.03 for Windows).

General Synthetic Experimental

^{1}H NMR spectra were recorded at 300K using a Bruker Avance DRX200, DRX300 or DPX 400 NMR spectrometer at a frequency of 200.1, 300.2 and 400.2 MHz respectively. ^{1}H NMR chemical shifts are reported in parts per million (ppm) and are referenced to solvent residual signals: $CDCl_3$ (δ 7.26), MeOD (δ 3.31), $(CD_3)_2CO$ (δ 2.05). ^{1}H NMR data is reported as chemical shift (δ_H), relative integral, multiplicity (s = singlet, d = doublet, t = triplet, q = quartet, dd = doublet of doublets, ddd = doublet of doublet of doublets), coupling constant (*J* Hz) and assignment where possible. ^{13}C NMR assignments were made in conjunction with DEPT experiments (C = quaternary carbon, CH = tertiary carbon, CH_2 = secondary carbon, CH_3 = primary carbon, C=O = carbonyl carbon). All 2D NMR experiments were carried out at 300K using a Bruker AVANCE DRX400 NMR spectrometer.

Low resolution mass spectra were recorded on a Finnigan LCQ Deca ion trap mass spectrometer (ESI). High resolution mass spectra were recorded on a Bruker 7T Fourier Transform Ion Cyclotron Resonance Mass Spectrometer (FTICR).

Melting points were recorded using a Stanford Research Systems OptiMelt Automated Melting Point System. Infrared (IR) absorption spectra were recorded on a Bruker ALPHA Spectrometer with Attenuated Total Reflection (ATR) capability, using OPUS 6.5 software.

Preparative reverse phase HPLC was performed using a Waters 600 Multisolvent Delivery System and Waters 500 pump with a Waters 2996 photodiode array detector or Waters 490E programmable wavelength detector operating at 254 and 280 nm. A Waters Sunfire 5 μm, 19 x 150 mm column was used at a flow rate of 7 mL min^{-1}. Preparative HPLC was performed utilising an at-column dilution loading scheme at a flow rate of 0.225 mL min^{-1}. Compounds were eluted with 0.1% TFA in water (solvent A), and 0.1% TFA in CH_3CN (solvent B) using a linear gradient of 0-40% B over 50 min.

LC-MS was performed on a Thermo Separation Products: Spectra System consisting of P400 Pump and a UV6000LP Photodiode array detector on a Phenomenex Jupiter 5 μm,

2.1 x 150 mm column at a flow rate of 0.2 mL min^{-1} coupled to a Thermoquest Finnigan LCQ Deca mass spectrometer (ESI) operating in positive mode. Separations involved a mobile phase of 0.1% formic acid in water (Solvent A) and 0.1% formic acid in acetonitrile (Solvent B) using a linear gradient of 0-100% B over 30 min.

Ion exchange was performed using Amberlite IR-120 (H^+) cation exchange resin which was prepared by washing with milli-Q water, 10% NaOH, milli-Q water, 10% HCl, and finally milli-Q water before use.

Materials

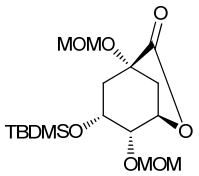
Analytical thin layer chromatography (TLC) was performed on commercially prepared silica plates (Merck Kieselgel 60 0.25 mm F254). Flash column chromatography was performed using 230-400 mesh Kieselgel 60 silica eluting with distilled solvents as described. Ratios of solvents used for TLC and column chromatography are expressed in v/v as specified. Compounds were visualised by UV light at 254 nm or using vanillin or cerium molybdate stain.

Commercial materials were used as received unless otherwise noted. DCM and methanol were distilled from calcium hydride, and THF and diethyl ether were distilled over sodium/benzophenone. Anhydrous DMF was purchased from Sigma Aldrich.

Experimental Procedures

Synthesis of ene-yne intermediate 3

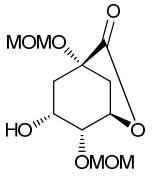
(1*R*, 3*R*, 4*S*, 5*R*)-3-(*tert*-butyldimethylsilyloxy)-1,4-bis(methoxymethoxy)-6-oxabicyclo[3.2.1]octan-7-one (7)



Phosphorus pentoxide (6.5 g, 23 mmol) was added every hour to a stirred mixture of 3-silylated lactone **6** (2.5 g, 8.3 mmol) and dimethoxymethane (100 mL, 1.1 mol) in dichloromethane (100 mL). The suspension was stirred vigorously for 2 h. Another portion of dimethoxymethane (40 mL, 0.4 mol) was then added and the reaction was stirred at rt for 1 h. The reaction mixture was then diluted with dichloromethane (125 mL) and washed with saturated aqueous $NaHCO_3$ solution (150 mL). The aqueous layer was further extracted with diethyl ether (3 × 100 mL) and the combined organic layers were dried over anhydrous Na_2SO_4 and the solvent was removed *in vacuo*. The crude residue was purified by flash column chromatography (eluent: 1:1 v/v diethyl ether/hexane) to afford lactone **7** as a pale yellow oil (4.6 g, 78%).

R_f (1:1 v/v diethyl ether/hexane) = 0.46; $[\alpha]_D^{25}$ -44° (*c* 1.25 in CH_2Cl_2); ν_{max}(ATR)/cm^{-1} 2954, 2931, 2896, 1789; ^{1}H NMR (400 MHz, $CDCl_3$): δ 4.84-4.67 (5H, m, 2×O*C*H_2CH$_3$ + H-5), 3.95-3.91 (2H, m, H-3 + H-4), 3.40 (3H, s, OCH$_2$*CH$_3$*), 3.32 (3H, s, OCH$_2$*CH$_3$*), 2.59-2.46 (2H, m, H-2$_{eq}$ + H-6$_{ax}$), 2.08-2.06 (2H, m, H-2$_{ax}$ + H-6$_{eq}$), 0.87 (9H, s, *Me$_3$*CSi), 0.06 (6H, s, 2*Me*Si); ^{13}C NMR (100 MHz, $CDCl_3$): δ 175.5 (C=O), 97.9 (CH_2), 92.9 (CH_2), 75.2 (C), 72.5 (CH), 67.7 (CH), 56.0 (CH_3), 55.8 (CH_3), 38.3 (CH_2), 35.3 (CH_2), 26.1 (CH_3), 18.4 (Me$_3$*C*Si), -4.3 (CH_3), -4.5 (CH_3). HRMS calcd. for $C_{17}H_{32}O_7SiNa$ (MNa^+) 399.1810; found 399.1812.

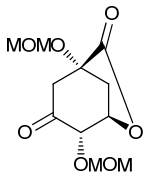
(1*S*,3*R*,4*R*,5*R*)-3-hydroxy-1,4-bis(methoxymethoxy)-6-oxabicyclo[3.2.1]octan-7-one (8)



TBAF (8.20 mL of a 1 M solution in THF, 8.20 mmol) was added dropwise to a solution of lactone **7** (2.6 g, 6.9 mmol) in THF (54 mL) at 0 ^{o}C. The reaction was stirred at rt for 2.5 h. The reaction mixture was then diluted with ethyl acetate (75 mL) and washed with saturated aqueous NH_4Cl solution (75 mL), brine (75 mL) and dried over anhydrous Na_2SO_4. The solvent was removed *in vacuo* and the crude residue was purified by flash column chromatography (eluent: 5:1 v/v diethyl ether/hexane) to afford alcohol **8** as a pale yellow oil (1.6 g, 87%).

R_f (5:1 v/v diethyl ether/hexane) = 0.21; $[\alpha]_D^{25}$ -32^{o} (*c* 1.5 in $CHCl_3$); ν_{max}(ATR)/cm^{-1} 3464, 2924, 2854, 1789; ^{1}H NMR (300 MHz, $CDCl_3$): δ 4.83-4.72 (5H, m, 2×O*C*$H_2$$CH_3$+ H-5), 3.98 (1H, dd, *J* 4.8, 4.8 Hz, H-4), 3.90 (1H, ddd, *J* 4.4, 6.4, 11.2 Hz, H-3), 3.42 (3H, s, OCH_2CH_3), 3.40 (3H, s, OCH_2*C*H_3), 2.49 (1H, d, *J* 11.4 Hz, H-6$_{ax}$), 2.57 (1H, ddd, *J* 2.7, 6.0, 11.4 Hz, H-6$_{eq}$), 2.32 (1H, ddd, *J* 2.7, 6.3, 11.4 Hz , H-2$_{eq}$), 1.88 (1H, t, *J* 11.4 Hz, H-2$_{ax}$); ^{13}C NMR (100 MHz, $CDCl_3$): δ 174.0 (C=O), 96.0 (CH_2), 93.0 (CH_2), 76.4 (C), 75.8 (C-H), 72.4 (C-H), 65.2 (CH), 56.1 (CH_3), 56.0 (CH_3), 35.5 (CH_2), 34.2 (CH_2).

(1*R*, 4*S*, 5*R*)-1,4-bis(methoxymethoxy)-6-oxabicyclo[3.2.1]octane-3,7-dione (9)

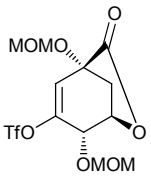


To a suspension of Dess-Martin Periodinane (2.7 g, 6.3 mmol) in anhydrous dichloromethane (13 mL) was added alcohol **8** (1.6 g, 6.0 mmol) in dichloromethane (13 mL). The reaction was allowed to stir at rt for 1.5 h. Diethyl ether (75 mL) was then added followed by a 1:1 (v/v) mixture of saturated aqueous $NaHCO_3$ solution and saturated aqueous $Na_2S_2O_3$ solution until all solid material had dissolved. The aqueous layer was extracted with diethyl ether (2 × 75 mL), the organic layer was dried over anhydrous Na_2SO_4 and the solvent removed *in vacuo* to afford ketone **9** as a pale yellow oil which was used without further purification (1.4 g, 90%).

R_f (3:1 v/v diethyl ether/hexane) = 0.58; $[\alpha]_D^{25}$ -56^{o} (*c* 7.3 in $CHCl_3$); ν_{max}(ATR)/cm^{-1} 2918, 1798, 1729; ^{1}H NMR (400 MHz, $CDCl_3$): δ 4.90-4.78 (4H, m, OC*H*$_2$CH$_3$ + OC*H*HCH$_3$ + H-5), 4.64 (1H, d, *J* 6.6 Hz, OCH*H*CH$_3$), 3.97 (1H, d, *J* 3.6 Hz, H-4), 3.41 (3H, s, OCH_3), 3.36 (3H, s, OCH_3), 3.05 (1H, d, *J* 17.4 Hz, H-2$_{ax}$), 2.92 (1H, ddd, *J* 3.2, 6.4, 12.0, H-6$_{eq}$), 2.84 (1H, dd, *J* 2.8, 17.2 Hz, H-2$_{eq}$), 2.68 (1H, d, *J* 12.0 Hz, H-6$_{ax}$); ^{13}C NMR (100 MHz,

$CDCl_3$): δ 201.9 (C=O), 173.8 (C=O), 96.4 (CH_2) , 93.2 (CH_2), 75.4 (C), 73.9 (C-H), 73.1 (C-H), 56.1 (CH_3), 56.1 (CH_3), 48.6 (CH_2), 33.9 (CH_2).

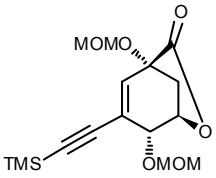
(1*R*, 4*S*, 5*R*)-1,4-bis(methoxymethoxy)-7-oxo-6-oxabicyclo[3.2.1]oct-2-en-3-ylttrifluoromethanesulfonate (10)



To a solution of KHMDS (4.1 mL of a 0.5 M solution in toluene, 2.07 mmol) in anhydrous DMF (2 mL) at -78 ^oC was added dropwise a solution of ketone **9** (414 mg, 1.6 mmol) in dry DMF (3.5 mL) and toluene (2.2 mL). The mixture was stirred for 1 h at -78 ^oC before a solution of 2-[*N*,*N*-bistrifluoromethylsulfonyl)amino]-5-chloropyridine (940 mg, 2.40 mmol) in anhydrous DMF (3.5 mL) was added dropwise. The resulting mixture was stirred at rt for 15 h at which point the reaction was diluted with ethyl acetate (100 mL) and washed with water (5 × 20 mL). The organic layers were dried over anhydrous Na_2SO_4 and solvent was removed *in vacuo*. The crude residue was then purified by flash column chromatography (eluent: 2:1 v/v diethyl ether/hexane) to afford vinyl triflate **10** as a pale yellow oil (415 mg, 67%).

R_f (2:1 v/v diethyl ether/hexane) = 0.48; $[\alpha]_D^{25}$ -83^o (*c* 1.0 in CH_2Cl_2); ν_{max}(ATR)/cm^{-1} 2958, 2904, 1807, 1664; ^{1}H NMR (400 MHz, $CDCl_3$): δ 6.30 (1H, d, *J* 1.3 Hz, H-2), 4.91 (1H, d, *J* 7.6 Hz, OC*H*HCH$_3$), 4.86 (1H, dd, *J* 3.7, 5.8 Hz, H-5), 4.80 (1H, d, *J* 7.0 Hz, OCH*H*CH$_3$), 4.78 (1H, d, *J* 8.6 Hz, OC*H*HCH$_3$), 4.76 (1H, d, *J* 7.0 Hz, OCH*H*CH$_3$), 4.27 (1H, d, *J* 3.4 Hz, H-4), 3.45 (3H, s, OCH$_3$), 3.43 (3H, s, OCH$_3$), 2.68 (1H, ddd, *J* 1.2, 6.0, 11.5 Hz, H-6$_{eq}$), 2.46 (1H, d, *J* 11.5 Hz, H-6$_{ax}$); ^{13}C NMR (100 MHz, $CDCl_3$): δ 172.3 (C=O), 145.0 (C), 125.3 (CH), 119.8 (q, $J_{C\text{-}F}$, 322 Hz, CF_3), 97.8 (CH_2), 93.9 (CH_2), 76.0 (C), 73.4 (CH), 72.3 (CH), 56.4 (CH_3), 56.4 (CH_3), 34.4 (CH_2); HRMS (APCI) m/z: calcd. for $C_{12}H_{16}F_3O_9S$ $[M+H]^+$: 393.0467, found 393.0462.

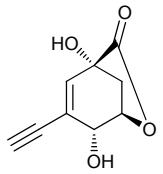
(1*R*,4*R*,5*R*)-1,4-bis(methoxymethoxy)-3-((trimethylsilyl)ethynyl)-6-oxabicyclo[3.2.1]oct-2-en-7-one (11)



Ethynyltrimethylsilane (0.75 mL, 5.3 mmol) and piperidine (1.60 mL, 15.9 mmol) were added to a suspension of vinyl triflate **10** (415 mg, 1.06 mmol), tetrakis(triphenylphosphine) palladium(0) (210 mg, 180 μmol) and copper iodide (40 mg, 210 μmol) in anhydrous THF (30 mL). The resulting solution was heated at 40 ^{o}C for 3.5 h before cooling to rt. Saturated aqueous NH_4Cl solution (75 mL) and diethyl ether (75 mL) were added and the organic layer was separated. The aqueous phase was extracted with diethyl ether (2 × 75 mL) and the combined organic extracts were dried over anhydrous Na_2SO_4 and the solvent was removed *in vacuo*. The crude residue was purified by flash column chromatography (eluent 1:1 v/v diethyl ether/hexane) to afford ene-yne **11** as a dark orange oil (280 mg, 75%).

R_f (1:1 v/v diethyl ether/hexane) = 0.33; $[\alpha]_D^{25}$ -134^{o} (*c* 2.3 in $CHCl_3$). ν_{max}(ATR)/cm^{-1} 2958, 2899, 1801; ^{1}H NMR (400 MHz, $CDCl_3$): δ 6.44 (1H, d, *J* 1.7 Hz, H-2), 4.87 (1H, d, *J* 7.5 Hz, OC*H*HCH$_3$), 4.85 (1H, d, *J* 7.0 Hz, OC*H*HCH$_3$), 4.79 (1H, d, *J* 7.6 Hz, OCH*H*CH$_3$), 4.74 (1H, d, *J* 6.8 Hz, OCH*H*CH$_3$), 4.72 (1H, dd, *J* 2.9, 6.1 Hz, H-5), 4.12 (1H, d, *J* 3.1 Hz, H-4), 3.42 (3H, s, OCH_3), 3.40 (3H, s OCH_3), 2.61 (1H, ddd, *J* 1.8, 6.1, 11.2 Hz, H-6$_{eq}$), 2.42 (1H, d, *J* 11.2 Hz, H-6$_{ax}$), 0.17 (9H, s, 3 × CH_3) ;^{13}C NMR (100 MHz, $CDCl_3$): δ 173.2 (C=O), 139.8 (CH), 122.0 (C), 101.5 (C), 97.5 (C), 97.0 (CH_2), 93.5 (CH_2), 77.5 (C), 74.2 (CH), 72.0 (CH), 56.3 (2 × CH_3), 34.4 (CH_2), -0.5 (CH_3); HRMS (APCI) m/z: calcd. for $C_{16}H_{25}O_6Si$ [*M*+H]$^+$: 341.1420, found 341.1415.

(1*R*,4*R*,5*R*)-3-ethynyl-1,4-dihydroxy-6-oxabicyclo[3.2.1]oct-2-en-7-one (3)



A solution of ene-yne **11** (76 mg, 0.22 mmol) in THF (9 mL) was treated with a solution of TBAF (74 μL of a 1.0 M in THF, 0.074 mmol) and the reaction mixture was stirred for 15 min at rt. The reaction mixture was diluted with ethyl acetate (50 mL), washed with saturated aqueous NH_4Cl (20 mL), brine (20 mL) and dried over anhydrous Na_2SO_4. The solvent was removed *in vacuo* to yield the intermediate protected ene-yne **12** which was used in the next step without purification. The protected ene-yne **12** was treated with 9:1 v/v trifluoroacetic acid/water (11 mL) for 30 min at 0 ^{o}C followed by stirring for a further 3 h at rt. At this point

the solvent was removed *in vacuo* to yield the crude residue which was purified by flash column chromatography to afford the ene-yne **3** (eluent 3:1 v/v diethyl ether/hexane) as a yellow oil (25 mg, 64%).

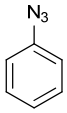
R_f (3:1 v/v diethyl ether/hexane) = 0.17. $[\alpha]_D^{25}$ -234^o (*c* 1.0 in MeOH). ν_{max}(ATR)/cm^{-1} 3293, 3283, 3270, 2356, 1785; ^{1}H NMR (400 MHz, CD_3OD): δ 6.35 (s, 1H, H-2), 4.66 (dd, 1H, *J* 5.6, 3.2 Hz, H-5), 4.09 (d, 1H, *J* 3.2 Hz, H-4), 3.48 (s, 1H, C*H*), 2.39 (ddd, 1H, *J* 11.2, 5.6, 1.6 Hz, H-6$_{eq}$), 2.34 (d, 1H, *J* 11.0 Hz, H-6$_{ax}$); The spectral data are in agreement with those reported by Prazeres *et al.*[6]

Synthesis of aryl and heteroaryl azides

General procedure A: Aromatic Azide Synthesis[7]

Aromatic aniline (1 eq.) was dissolved in dry CH_3CN (10 mL) and the solution was cooled to 0 ^{o}C in an ice bath. To this mixture was added *tert*-butylnitrite (1.5 eq.) followed by dropwise addition of azidotrimethylsilane (1.2 eq.). The reaction mixture was allowed to warm to rt and stirred for a further 1 h. The solvent was removed *in vacuo* and the product was purified by flash column chromatography to afford the corresponding azide.

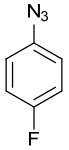
Azidobenzene[7]



Aniline (500 mg, 5.4 mmol, 0.48 mL), dry CH_3CN (10 mL), *tert*-butylnitrite (8 mmol, 0.96 mL) and azidotrimethylsilane (6.4 mmol, 0.85 mL) were reacted under general procedure A. The crude compound was purified by flash column chromatography (eluent: 9:1 v/v ethyl acetate/hexane) to afford azidobenzene as a yellow oil (450 mg, 70%).

R_f (9:1 v/v ethyl acetate /hexane) = 0.22; ν_{max}(ATR)/cm^{-1} 2921, 2853, 2118; ^{1}H NMR (400 MHz, $CDCl_3$): δ 7.37 (2H, ddd, *J* 0.8, 1.2, 7.6 Hz, Ar-H, C-3 + C-5), 7.17 (2H, ddd, *J* 0.8, 1.2, 7.6 Hz, Ar-H, C-2 + C-6), 7.04 (1H, ddd, *J* 0.8, 1.2, 7.6 Hz, Ar-H, C-4). LRMS (APCI+): [*M*+H$^+$] 121.1. These data are in agreement with those reported by Barral *et al.*[7]

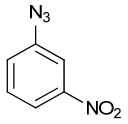
1-azido-4-fluorobenzene[7]



4-fluoroaniline (0.43 mL, 4.5 mmol), dry CH_3CN (10 mL), *tert*-butylnitrite (0.80 mL, 6.8 mmol) and azidotrimethylsilane (0.72 mL, 5.3 mmol) were reacted under general procedure A. The crude compound was purified by flash column chromatography (eluent: 4:1 v/v ethyl acetate/hexane) to afford 1-azido-4-fluorobenzene as an orange oil (430 mg, 70%).

R_f (4:1 v/v ethyl acetate/hexane) = 0.79; ν_{max}(ATR)/cm^{-1} 2923, 2854, 2110, 2066. $^1H(^{19}F)$ NMR (300 MHz, $CDCl_3$): δ 7.06 (2H, d, J 8.7 Hz, Ar-H, C-2 + C-6); 6.99 (2H, d, *J* 9.0 Hz, Ar-H, C-3 + C-5). These data are in agreement with those reported by Barral *et al.*[7]

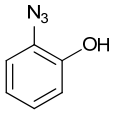
1-azido-3-nitrobenzene[7]



3-nitroaniline (500 mg, 3.6 mmol), dry CH_3CN (10 mL), *tert*-butylnitrite (0.64 mL, 5.4 mmol) and azidotrimethylsilane (0.57 mL, 4.3 mmol) were reacted under general procedure A. The crude compound was purified by flash column chromatography (eluent: 4:1 v/v ethyl acetate/hexane) to afford 1-azido-3-nitrobenzene as an orange crystalline solid (490 mg, 82%).

R_f(4:1 v/v ethyl acetate/hexane) = 0.3; mp 59-60 ^oC; ν_{max}(ATR)/cm^{-1} 3095, 2121; ^1H NMR (300 MHz, $CDCl_3$): δ 7.99 (1H, ddd, *J* 1.2, 2.0, 8.0 Hz, H-4), 7.90 (1H, dd, *J* 2.0, 2.0 Hz, H-2), 7.54 (1H, dd, *J* 8.0, 8.0 Hz, H-5), 7.34 (1H, ddd, *J* 1.3, 2.0, 8.0 Hz, H-6); ^{13}C NMR (100 MHz, $CDCl_3$): δ 149.3 (C), 142.0 (C), 130.5 (CH), 125.0 (CH), 119.7 (CH), 114.2 (CH). These data are in agreement with those reported by Barral *et al.*[7]

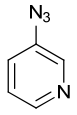
2-azidophenol[7]



2-aminophenol (500 mg, 4.60 mmol), dry CH_3CN (10 mL), *tert*-butylnitrite (0.80 mL, 6.8 mmol) and azidotrimethylsilane (0.68 mL, 5.1 mmol) were reacted under general procedure A. The crude compound was purified by flash column chromatography (eluent: 4:1 v/v ethyl acetate/hexane) to afford 2-azidophenol as a dark red oil (440 mg, 70%).

R_f (4:1 v/v ethyl acetate /hexane) = 0.34; ν_{max}(ATR)/cm^{-1} 3437, 2924, 2125; ^{1}H NMR (400 MHz, $CDCl_3$): δ 7.07 (2H, dd, *J* 1.2, 7.6 Hz, Ar-H, C-3 + C-6), 6.94 (2H, t, *J* 7.8 Hz, Ar-H, C-4 + C-5); LRMS $[M\text{-H}]^-$ 134.0. These data are in agreement with those reported by Barral *et al.*[7]

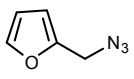
3-azidopyridine[8]



A solution of sodium nitrite (460 mg, 6.4 mmol) in water (1.2 mL) was added dropwise over 10 min to a stirred solution of 3-aminopyridine (506 mg, 5.3 mmol) in 10% HCl (4.5 mL) at 0 ^oC. After stirring for 15 min, a solution of sodium azide (370 mg, 5.6 mmol) in water (1.2 mL) was added dropwise over 5 min. The reaction mixture was stirred for an additional 45 min at room temperature before the reaction was quenched by the addition of saturated aqueous $NaHCO_3$ solution (10 mL). The reaction mixture was extracted with $CHCl_3$ (3 × 15 mL) and the combined organic fractions washed with water (40 mL), dried over anhydrous Na_2SO_4 and the solvent was removed *in vacuo*. The crude residue was purified by flash column chromatography (eluent: 4:1 v/v ethyl acetate/hexane) to yield 3-azidopyridine as a dark red oil (520 mg, 82%).

R_f (4:1 v/v ethyl acetate/hexane) = 0.28. ν_{max}(ATR)/cm^{-1} 3034, 2428, 2273, 2088. ^{1}H NMR (300 MHz, $CDCl_3$): δ 8.41-8.37 (2H, m, Ar-H, C-2 + C-6), 7.38-7.27 (2H, m, Ar-H, C-4 + C-5). These data are in agreement with those reported by Crabtree *et al*.[8]

2-(azidomethyl)furan[9]

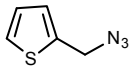


DBU (0.60 mL, 4.1 mmol) was added dropwise to a solution of furfuryl alcohol (0.38 mL, 5.0 mmol) and diphenylphosphoryl azide (1.3 mL, 6.0 mmol) in anhydrous toluene (15 mL)

at 0 ^{o}C. The reaction mixture was stirred for 16 h at rt. The reaction mixture was washed with water (10 mL), dried over anhydrous Na_2SO_4 and the solvent removed *in vacuo* (60 mbar). The resulting residue was purified by flash column chromatography (eluent: 9:1 v/v ethyl acetate/hexane) to afford 2-(azidomethyl)furan as a colourless oil (250 mg, 41%).

R_f (9:1 v/v hexane/ethyl acetate) = 0.35; ν_{max}(ATR)/cm^{-1} 2924, 2854, 2171, 2099; ^{1}H NMR (400 MHz, $CDCl_3$): δ 7.43 (1H, dd, *J* 0.8, 1.6 Hz, H-5), 6.37-6.34 (2H, m, H-3 + H-4), 4.29 (2H, s, CH_2). These data are in agreement with those previously by Rogers *et al.*[9]

2-(azidomethyl)thiophene[9]

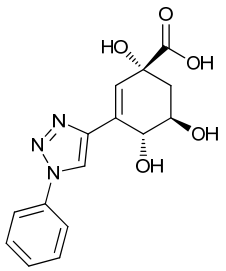


Imidazole-1-sulfonyl azide hydrochloride[10] (1.1 g, 5.3 mmol) was added to a suspension of 2-thiophenemethylamine (0.45 mL, 4.4 mmol), potassium carbonate (1.8 g, 13 mmol) and copper(II) sulfate pentahydrate (9.3 mg, 0.05 mmol) in methanol (10 mL). The reaction mixture was stirred at rt for 24 h. The mixture was diluted with water (25 mL), acidified to pH 1 *via* the dropwise addition of 1 M aqueous HCl and extracted with ethyl acetate (3 × 25 mL). The combined organic fractions were washed with aqueous 1 M HCl (50 mL), dried over anhydrous Na_2SO_4 and concentrated *in vacuo* (60 mbar). The crude residue was purified by flash column chromatography (eluent: 2:1 v/v ethyl acetate/hexane) to afford 2-(azidomethyl)thiophene as a colourless oil (250 mg, 41%).

R_f (2:1 v/v ethyl acetate/hexane) = 0.84; ν_{max}(ATR)/cm^{-1} 2088; ^{1}H NMR (400 MHz, $CDCl_3$): δ 7.30 (1H, dd, *J* 1.2, 5.2 Hz, H-5), 7.04-6.98 (2H, m, H-3 + H-4), 4.46 (2H, s, CH_2). These data are in agreement with those previously reported by Rogers *et al.*[9]

Synthesis of triazole-based inhibitors 4a-f, 5a and 5b

(1*R*, 4*R*, 5*R*)-1,4,5-trihydroxy-3-(1-phenyl-1*H*-1,2,3-triazol-4-yl)cyclohex-2-enecarboxylic acid (4a)

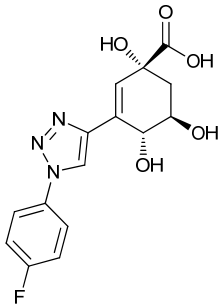


A freshly prepared 1.0 M solution of aqueous sodium ascorbate (8.8 μL, 8.8 μmol) and 0.32 M aqueous copper(II) sulfate (2.8 μL, 0.88 μmol) solution were added sequentially to a suspension of **3** (7.9 mg, 0.044 mmol) and azidobenzene (10 mg, 0.090 mmol) in a mixture of *tert*-butanol/water (1:1 v/v, 0.36 mL). The resulting heterogeneous reaction mixture was stirred vigorously for 24 h at rt. The reaction mixture was then diluted with ethyl acetate (10 mL) and water (10 mL) and the aqueous phase extracted with ethyl acetate (2 × 20 mL). The combined organic layers were dried over anhydrous Na_2SO_4 and the solvent removed *in vacuo*. The crude residue was purified by flash column chromatography (eluent: 9:1 v/v dichloromethane/methanol) to yield the triazole lactone as a yellow oil. Lithium hydroxide (180 μL of a 0.5 M aqueous solution, 0.090 mmol) was added to a solution of the triazole lactone (10 mg, 0.040 mmol) in THF (320 μL) and the reaction was stirred at rt for 1 h. The solvent was removed *in vacuo* and the reaction mixture was diluted with water (2 mL) and washed with ethyl acetate (2 × 2 mL). The aqueous layer was then treated with Amberlite IR-120 (H^+ form) until pH 6 was reached. The resin was filtered and washed with milli-Q water. The aqueous fraction was lyophilised to afford the desired acid **4a** as an off-white solid (7.8 mg, 56% over two steps).

mp 218-219 ^oC (decomp.); $[\alpha]_D^{25}$ -32^o (*c* 2.4 in H_2O); ν_{max}(ATR)/cm^{-1} 3371, 3284, 1606; ^1H NMR (400 MHz, D_2O): δ 8.50 (1H, s, N-C*H*=C), 8.16 (1H, s, Ar-H, C-2), 8.01 (1H, d, *J* 8.0 Hz, Ar-H, C-4), 7.90 (1H, d, *J* 8.4 Hz, Ar-H, C-6), 7.61 (1H, dd, *J* 7.6, 8.0 Hz, Ar-H, C-5), 6.51 (1H, s, H-2), 4.46 (1H, d, *J* 7.2 Hz, H-4), 4.11 (1H, ddd, *J* 3.6, 6.8, 10.4 Hz, H-5), 2.23 (1H, dd, *J* 9.6, 14.0 Hz, H-6_{ax}), 2.18 (1H, dd, *J* 4.0, 14.0 Hz, H-6_{eq}); ^{13}C NMR (100 MHz, D_2O): δ 180.2, 145.5, 136.3, 130.8, 129.9, 129.3, 128.9, 121.9, 121.0, 73.5, 70.7, 69.9,

37.5; LC-MS [M+H^+] 318.0, R_t = 15.15 min; HRMS (ESI) m/z: calcd. for $C_{15}H_{16}N_3O_5$ [M+H^+]: 318.1084, found 318.1086.

(1*R*,4*R*,5*R*)-3-(1-(4-fluorophenyl)-1*H*-1,2,3-triazol-4-yl)-1,4,5-trihydroxycyclohex-2-enecarboxylic acid (4b)

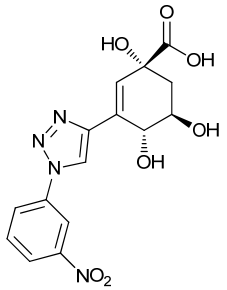


A freshly prepared 1.0 M solution of aqueous sodium ascorbate (45 μL, 45 μmol) and 0.32 M aqueous copper(II) sulfate solution (14 μL, 4.5 μmol) were sequentially added to a suspension of **3** (6.5 mg, 0.036 mmol) and 1-azido-4-fluorobenzene (14.4 mg, 0.10 mmol) in a mixture of *tert*-butanol/water (1:1 v/v, 0.36 mL). The resulting heterogeneous reaction mixture was stirred for 24 h at rt. The reaction mixture was then diluted with ethyl acetate (10 mL) and water (10 mL) and the aqueous phase extracted with ethyl acetate (2 × 20 mL). The combined organic layers were dried over anhydrous Na_2SO_4 and the solvent removed *in vacuo*. The crude residue was purified by flash column chromatography (eluent: 9:1 v/v dichloromethane/methanol) to yield the triazole lactone as a yellow oil. Lithium hydroxide (85 μL of a 0.5 M aqueous solution, 0.042 mmol) was added to a solution of the triazole lactone (6.4 mg, 0.019 mmol) in THF (155 μL) and the reaction was stirred at rt for 1 h. The solvent was removed *in vacuo* and the reaction mixture was diluted with water (2 mL) and washed with ethyl acetate (2 × 2 mL). The aqueous layer was then treated with Amberlite IR-120 (H^+ form) until pH 6 was reached. The resin was filtered and washed with milli-Q water. The aqueous fraction was lyophilised to afford the desired acid **4b** as an off-white solid (6.5 mg, 55% over two steps).

mp 182-183 ^oC (decomp.); $[\alpha]_D^{25}$ -26^o (*c* 2.5 in H_2O); ν_{max}(ATR)/cm^{-1} 3371, 3284, 1606; $^1H(^{19}F)$ NMR (300 MHz, D_2O): δ 8.52 (1H, s, N-C*H*=C), 7.71 (2H, d, *J* 9.0 Hz, Ar-H), 7.29 (2H, d, *J* 9.0 Hz, Ar-H), 6.42 (1H, s, H-2), 4.52 (1H, d, *J* 6.4 Hz, H-4), 4.07 (1H, ddd, *J* 4.1, 6.5, 10.3 Hz, H-5), 2.23 (1H, dd, *J* 9.6, 14.0 Hz, H-6_{ax}), 2.18 (1H, dd, *J* 4.0, 14.0 Hz, H-6_{eq}); ^{13}C NMR (100 MHz, D_2O): δ 180.5 (C=O), 162.6 (d, $J_{C\text{-}F}$ 246 Hz, *i*-Ar-C), 145.5, 132.6,

130.7, 128.7, 123.5 (d, $J_{C\text{-}F}$ 9 Hz, *m*-Ar-C), 122.3, 116.6 (d, $J_{C\text{-}F}$ 23 Hz, *o*-Ar-C), 70.8, 69.9, 37.6; LC-MS [*M*+H^+] 336.0, R_t = 15.94 min; HRMS (ESI) m/z: calcd. for $C_{15}H_{15}FN_3O_5$ [*M*+H^+]: 336.0990, found 336.0996.

(1*R*,4*R*,5*R*)-1,4,5-trihydroxy-3-(1-(3-nitrophenyl)-1*H*-1,2,3-triazol-4-yl)cyclohex-2-enecarboxylic acid (4c)

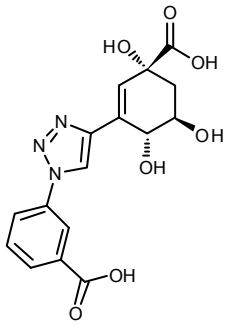


A freshly prepared 1.0 M solution of aqueous sodium ascorbate (44 μL, 44 μmol) and 0.32 M aqueous copper(II) sulfate solution (14 μL, 4.4 μmol) were added sequentially to a suspension of **3** (7.9 mg, 0.044 mmol) and 1-azido-3-nitrobenzene (10.8 mg, 0.07 mmol) in a mixture of *tert*-butanol/water (1:1 v/v, 0.36 mL). The resulting heterogeneous reaction mixture was stirred vigorously for 24 h at rt. The reaction mixture was then diluted with ethyl acetate (10 mL) and water (10 mL) and the aqueous phase extracted with ethyl acetate (2 × 20 mL). The combined organic layers were dried over anhydrous Na_2SO_4 and the solvent removed *in vacuo*. The crude residue was purified by flash column chromatography (eluent: 9:1 v/v dichloromethane/methanol) to yield the triazole lactone as a yellow oil. Lithium hydroxide (67 μL of a 0.5 M aqueous solution, 0.034 mmol) was added to a solution of the triazole lactone (4.6 mg, 0.013 mmol) in THF (250 μL) and the reaction was stirred at rt for 2 h. The solvent was removed *in vacuo* and the reaction mixture was diluted with water (2 mL). The solution was then treated with Amberlite IR-120 (H^+ form) until pH 6 was reached. The resin was filtered and washed with milli-Q water. The aqueous fraction was lyophilised to afford the desired acid **4c** as an off-white solid (4.9 mg, 30% over two steps).

mp 208-209 ^oC (decomp.); $[\alpha]_D^{25}$ -24^o (*c* 0.3 in H_2O); ν_{max}(ATR)/cm^{-1} 3333, 1644; ^1H NMR (400 MHz, D_2O): δ 8.69-8.68 (2H, m, N-C*H*=C + Ar-H C-4), 8.38 (1H, dd, *J* 1.2, 8.0 Hz, Ar-H C-2), 8.21 (1H, dd, *J* 1.6, 8.0 Hz, Ar-H, Ar-H, C-6), 7.84 (1H, dd, *J* 8.0, 8.0 Hz, Ar-H, C-5), 6.45 (1H, s, H-2), 4.52 (1H, d, *J* 6.8 Hz, H-4), 4.08 (1H, ddd, *J* 4.0, 6.4, 10.4 Hz,

H-5), 2.14 (1H, dd, *J* 10.0, 14.0 Hz, H-6_{ax}), 2.09 (1H, dd, *J* 4.0, 14.0 Hz, H-6_{eq}); ^{13}C NMR (100 MHz, D_2O): δ 180.2, 148.5, 145.8, 137.0, 133.3, 130.8, 128.8, 127.1, 123.0, 122.0, 116.1, 73.5, 71.0, 69.9, 37.7; LC-MS [M+H^+] 362.9, R_t = 15.95 min ; HRMS (ESI) m/z: calcd. for $C_{15}H_{14}N_4O_7Na$ [M+Na^+]: 385.0760, found 385.0755.

3-(4-((3*R*,5*R*,6*R*)-3-carboxy-3,5,6-trihydroxycyclohex-1-enyl)-1*H*-1,2,3-triazol-1-yl)benzoic acid (4d)

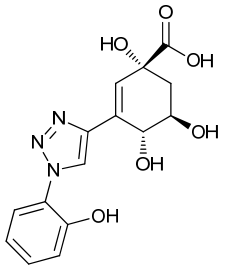


A freshly prepared 1.0 M solution of aqueous sodium ascorbate (4.2 μL, 4.2 μmol) and 0.32 M of aqueous copper(II) sulfate solution (1.3 μL, 0.42 μmol) were added sequentially to a suspension of **3** (7.5 mg, 0.042 mmol) and 3-azidobenzoic acid (8.0 mg, 0.050 mmol) in a mixture of *tert*-butanol/water (1:1 v/v, 0.36 mL). The resulting heterogeneous reaction mixture was stirred vigorously for 24 h at rt. The reaction mixture was then diluted with ethyl acetate (10 mL) and water (10 mL) and the aqueous phase extracted with ethyl acetate (2 × 20 mL). The combined organic layers were dried over anhydrous Na_2SO_4 and the solvent removed *in vacuo*. The crude residue was purified by flash column chromatography (eluent: 9:1 v/v dichloromethane/methanol) to yield the triazole lactone as a yellow oil. Lithium hydroxide (550 μL of a 0.5 M aqueous solution, 0.276 mmol) was added to a solution of triazole lactone (11 mg, 0.030 mmol) in THF (320 μL) and the reaction was stirred at rt for 24 h. The solvent was removed *in vacuo* before dilution with water (2 mL) and subsequent purification by preparative HPLC. The HPLC fractions containing the desired product were lyophilised to afford the desired diacid **4d** as a white, fluffy solid (4.1 mg, 27% over two steps).

mp 177-178 ^{o}C (decomp.); $[\alpha]_D^{25}$ -36^{o} (*c* 0.4 in H_2O); ν_{max}(ATR)/cm^{-1} 3328, 3279, 1710, 1594; ^{1}H NMR (400 MHz, D_2O): δ 8.52 (1H, s, N-C*H*=C), 8.21 (1H, s, Ar-H, C-2), 8.05 (1H, d, *J* 8.0 Hz, Ar-H, C-4), 7.93 (1H, d, *J* 8.4 Hz, Ar-H, C-6), 7.61 (1H, dd, *J* 7.6, 8.0

Hz, Ar-H, C-5), 6.52 (1H, s, H-2), 4.52 (1H, d, *J* 7.2 Hz, H-4), 4.12 (1H, m, H-5), 2.36-2.23 (2H, m, H-6_{ax}+ H-6_{eq}). ^{13}C NMR (100 MHz, D_2O): δ 178.2 (C=O), 169.4 (C=O), 145.2, 136.3, 132.2, 131.9, 130.3, 130.2, 127.0, 125.2, 122.0, 121.5, 71.1, 69.6, 37.8; LC-MS $[M+H]^+$ 362.0, R_t = 11.74 min; HRMS (ESI) m/z: calcd. for $C_{16}H_{15}N_3O_7Na$ [*M*+Na^+]: 384.0802, found 384.0802.

(1*R*,4*R*,5*R*)-1,4,5-trihydroxy-3-(1-(2-hydroxyphenyl)-1*H*-1,2,3-triazol-4-yl)cyclohex-2-enecarboxylic acid (4e)

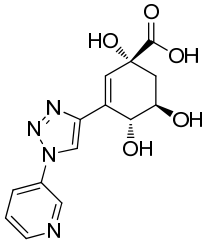


A freshly prepared 1.0 M solution of aqueous sodium ascorbate (45 μL, 45 μmol) and 0.32 M aqueous copper(II) sulfate solution (14 μL, 4.5 μmol) were added sequentially to a suspension of **3** (8.0 mg, 0.050 mmol) and 2-azidophenol (18 mg, 0.14 mmol) in a mixture of *tert*-butanol/water (1:1 v/v, 0.36 mL). The resulting heterogeneous reaction mixture was stirred vigorously for 24 h at rt. The reaction mixture was then diluted with ethyl acetate (10 mL) and water (10 mL) and the aqueous phase extracted with ethyl acetate (2 × 20 mL). The combined organic layers were dried over anhydrous Na_2SO_4 and the solvent removed *in vacuo*. The crude residue was purified by flash column chromatography (eluent: 9:1 v/v dichloromethane/methanol) to yield the triazole lactone as a yellow oil. Lithium hydroxide (115 μL of a 0.5 M aqueous solution, 0.058 mmol) was added to a solution of the triazole lactone (8.0 mg, 0.024 mmol) in THF (210 μL) and the reaction was stirred at rt for 1 h. The solvent was removed *in vacuo* and the reaction mixture was diluted with water (2 mL) and washed with ethyl acetate (2 × 2 mL). The aqueous layer was then treated with Amberlite IR-120 (H^+ form) until pH 6 was reached. The resin was filtered and washed with milli-Q water. The aqueous fraction was lyophilised to afford the desired acid **4e** as a dark brown solid (8.1 mg, 54% over two steps).

mp 84-85 ^{o}C (decomp.); $[\alpha]_D^{25}$ -31^o (*c* 2.2 in H_2O); ν_{max}(ATR)/cm^{-1} 3482, 3348, 1651; ^{1}H NMR (400 MHz, D_2O): δ 8.33 (1H, s, N-C*H*=C), 7.42 (1H, d, *J* 8.0 Hz, Ar-H, C-6), 7.34

(1H, t, *J* 8.0 Hz, Ar-H, C-5), 7.03 (1H, d, *J* 8.0 Hz, Ar-H, C-3), 6.97 (1H, t, *J* 8.0 Hz, Ar-H, C-4), 6.32 (1H, s, H-2), 4.42 (1H, d, *J* 6.8 Hz, H-4), 3.99 (1H, ddd, *J* 4.0, 6.4, 10.4 Hz, H-5), 2.13 (1H, dd, *J* 10.0, 14.0 Hz, H-6_{ax}), 2.08 (1H, dd, *J* 4.0, 13.6 Hz, H-6_{eq}); ^{13}C NMR (100 MHz, D_2O): δ 181.2 (C=O), 152.7, 144.4, 133.4, 130.9, 128.4, 126.1, 125.2, 118.7, 73.6, 71.3, 70.0, 37.4; LC-MS [*M*+H^+] 334.0, R_t = 13.85 min; HRMS (ESI) m/z: calcd. for $C_{15}H_{15}N_3O_6Na$ [*M*+Na^+]: 356.0853, found 356.0848.

(1*R*,4*R*,5*R*)-1,4,5-trihydroxy-3-(1-(pyridine-3-yl)-1H-1,2,3-triazol-4-yl)cyclohex-2-enecarboxylic acid (4f)

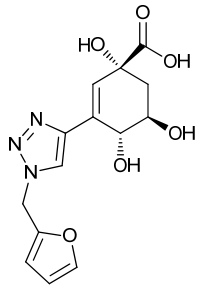


A freshly prepared 1.0 M solution of aqueous sodium ascorbate (49 μL, 49 μmol) and 0.32 M aqueous copper(II) sulfate solution (15 μL, 4.8 μmol) were added to a suspension of **3** (8.8 mg, 0.049 mmol) and 3-azidopyridine (11.8 mg, 0.10 mmol) in a mixture of *tert*-butanol/water (1:1 v/v, 0.36 mL). The resulting heterogeneous reaction mixture was stirred vigorously for 24 h at rt. The reaction mixture was then diluted with ethyl acetate (10 mL) and water (10 mL) and the aqueous phase extracted with ethyl acetate (2 × 20 mL). The combined organic layers were dried over anhydrous Na_2SO_4 and the solvent removed *in vacuo*. The crude residue was purified by flash column chromatography (eluent: 9:1 v/v dichloromethane/methanol) to yield the triazole lactone as a yellow oil. Lithium hydroxide (74 μL of a 0.5 M aqueous solution, 0.040 mmol) was added to a solution of the lactone (5.6 mg, 0.019 mmol) in THF (250 μL) and the reaction was stirred at rt for 2 h. The solvent was removed *in vacuo* and the reaction mixture was diluted with water (2 mL). The solution was then treated with Amberlite IR-120 (H^+ form) until pH 6 was reached. The resin was filtered and washed with milli-Q water. The aqueous fraction was lyophilised to afford the desired acid **4f** as a yellow solid (5.9 mg, 39% over two steps).

mp 189-190 ^{o}C (decomp.); $[\alpha]_D^{25}$ -34^{o} (*c* 0.4 in H_2O); ν_{max}(ATR)/cm^{-1} 3385, 1608; ^{1}H NMR (400 MHz, D_2O): δ 9.01 (1H, s, Ar-H, C-2), 8.68-8.64 (2H, m, Ar-H, C-6 + N-C*H*=C), 8.28 (1H, dd, *J* 2.4, 8.4 Hz, Ar-H, C-4), 7.69 (1H, dd, *J* 4.8, 8.4 Hz, Ar-H, C-5), 6.44 (1H, s,

H-2), 4.52 (1H, d, *J* 6.8 Hz, H-4), 4.07 (1H, ddd, *J* 4.4, 6.8, 9.6 Hz, H-5), 2.21 (1H, dd, *J* 9.6, 13.6 Hz, H-6_{ax}), 2.14 (1H, dd, *J* 4.0, 13.6 Hz, H-6_{eq}); ^{13}C NMR (100 MHz, D_2O): δ 180.4, 149.5, 145.8, 141.4, 130.6, 130.0, 128.9, 125.1, 122.1, 73.6, 71.0, 69.9, 37.7; LC-MS [M+H^+] 318.9, R_t = 12.80 min; HRMS (ESI) m/z: calcd. for $C_{14}H_{14}N_4O_5Na$ [M+Na^+]: 341.0862, found 341.0860.

(1*R*,4*R*,5*R*)-3-(1-(furan-2-ylmethyl)-1H-1,2,3-triazol-4-yl)-1,4,5-trihydroxycyclohex-2-enecarboxylic acid (5a)

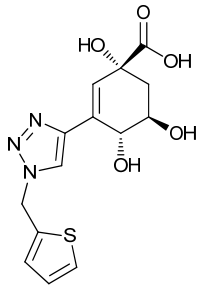


A freshly prepared 1.0 M solution of aqueous sodium ascorbate (45 μL, 45 μmol) and 0.32 M aqueous copper(II) sulfate solution (14 μL, 4.5 μmol) were added sequentially to a suspension of **3** (8.1 mg, 0.045 mmol) and 2-azidomethylfuran (14.4 mg, 0.117 mmol) in a mixture of *tert*-butanol/water (1:1 v/v, 0.36 mL). The resulting heterogeneous reaction mixture was stirred vigorously for 24 h at rt. The reaction mixture was then diluted with ethyl acetate (10 mL) and water (10 mL) and the aqueous phase extracted with ethyl acetate (2 × 20 mL). The combined organic layers were dried over anhydrous Na_2SO_4 and solvent removed *in vacuo*. The crude residue was purified by flash column chromatography (eluent: 9:1 v/v dichloromethane/methanol) to yield the triazole lactone as a yellow oil. Lithium hydroxide (155 μL of a 0.5 M aqueous solution, 0.077 mmol) was added to a solution of the triazole lactone (9.5 mg, 0.031 mmol) in THF (320 μL) and the reaction was stirred at rt for 1 h. The solvent was removed *in vacuo* and the reaction mixture was diluted with water (2 mL) and washed with ethyl acetate (2 × 2 mL). The aqueous layer was then treated with Amberlite IR-120 (H^+ form) until pH 6 was reached. The resin was filtered and washed with milli-Q water. The aqueous fraction was lyophilised to afford the desired acid **5a** as an off-white solid (9.5 mg, 69% over two steps).

mp 188-189 ^{o}C (decomp.); $[\alpha]_D^{25}$ -29^{o} (*c* 1.0 in H_2O); ν_{max}(ATR)/cm^{-1} 3128,1589; ^{1}H NMR (400 MHz, D_2O): δ 8.17 (1H, s, N-C*H*=C), 7.53 (1H, d, *J* 1.6 Hz, Ar-H, C-5), 6.60 (1H, d, *J* 3.6 Hz, Ar-H, C-3), 7.07 (1H, dd, *J* 1.6, 3.2 Hz, Ar-H, C-4), 6.41 (1H, s, H-2), 5.66

(2H, s, CH_2), 4.44 (1H, d, *J* 6.8 Hz, H-4), 4.08 (1H, ddd, *J* 3.6, 6.8, 10.4 Hz, H-5), 2.26 (1H, dd, *J* 10.4, 13.2 Hz, H-6_{ax}), 2.16 (1H, dd, *J* 3.6, 14.0 Hz, H-6_{eq}); ^{13}C NMR (100 MHz, D_2O): δ 178.0 (C=O), 147.4, 144.1, 132.1, 126.4, 123.8, 110.8, 110.4, 72.9, 71.0, 69.5, 46.8, 37.4; LC-MS [*M*+H^+] 322.0, R_t = 10.62 min; HRMS (ESI) m/z: calcd. for $C_{14}H_{16}N_3O_6$ [*M*+H^+]: 322.1033, found 322.1033.

(1*R*,4*R*,5*R*)-1,4,5-trihydroxy-3-(1-(thiophen-2-ylmethyl)-1*H*-1,2,3-triazol-4 yl)cyclohex-2-enecarboxylic acid (5b)



A freshly prepared 1.0 M solution of aqueous sodium ascorbate (47 μL, 47 μmol) and 0.32 M aqueous copper(II) sulfate solution (15 μL, 4.7 μmol) were added sequentially to a suspension of **3** (8.4 mg, 0.047 mmol) and 2-azidomethylthiophene (16.1 mg, 0.120 mmol) in a mixture of *tert*-butanol/water (1:1 v/v, 0.36 mL). The resulting heterogeneous reaction mixture was stirred vigorously for 24 h at rt. The reaction mixture was then diluted with ethyl acetate (10 mL) and water (10 mL) and the aqueous phase extracted with ethyl acetate (2 × 20 mL). The combined organic layers were dried over anhydrous Na_2SO_4 and the solvent removed *in vacuo*. The crude residue was purified by flash column chromatography (eluent: 9:1 v/v dichloromethane/methanol) to yield the triazole lactone as a yellow oil. Lithium hydroxide (200 μL of a 0.5 M aqueous solution, 0.10 mmol) was added to a solution of the triazole lactone (10.8 mg, 0.034 mmol) in THF (320 μL) and the reaction was stirred at rt for 1 h. The solvent was removed *in vacuo* and the reaction mixture was diluted with water (2 mL) and washed with dichloromethane (2 × 2 mL). The aqueous layer was then treated with Amberlite IR-120 (H^+ form) until pH 6 was reached. The resin was filtered and washed with milli-Q water. The aqueous fraction was lyophilised to afford the desired acid **5b** as an off-white solid (10.9 mg, 69% over two steps).

mp 145-146 ^{o}C (decomp.); $[\alpha]_D^{25}$ -39^{o} (*c* 0.4 in H_2O); ν_{max}(ATR)/cm^{-1} = 3275, 1641; ^{1}H NMR (400 MHz, D_2O): δ 8.17 (1H, s, N-C*H*=C), 7.47 (1H, dd, *J* 1.2, 5.2 Hz, Ar-H, C-3), 7.23 (1H, dd, *J* 0.8, 3.6 Hz, Ar-H, C-5), 7.07 (1H, dd, *J* 3.6, 5.2 Hz, Ar-H, C-4), 6.42 (1H, s,

H-2), 5.83 (2H, s, CH_2), 4.41 (1H, d, *J* 7.2 Hz, H-4), 4.08 (1H, ddd, *J* 4.0, 7.2, 10.8 Hz, H-5), 2.26 (1H, dd, *J* 10.4, 13.2 Hz, H-6_{ax}), 2.08 (1H, dd, *J* 4.8, 13.6 Hz, H-6_{eq}); ^{13}C NMR (100 MHz, D_2O): δ 177.5 (C=O), 144.4, 136.3, 132.5, 128.5, 127.6, 127.5, 125.8, 123.7, 72.7, 71.1, 69.4, 48.4, 37.7; LC-MS [*M*+H^+] 338.0, R_t = 12.08 min; HRMS (ESI) m/z: calcd. for $C_{14}H_{16}N_3O_5S$ [*M*+H^+]: 338.0805, found 338.0807.

Glide Docking solutions of 4a-f, 5a and 5b against *S. coelicolor* and *H. pylori* type II dehydroquinase

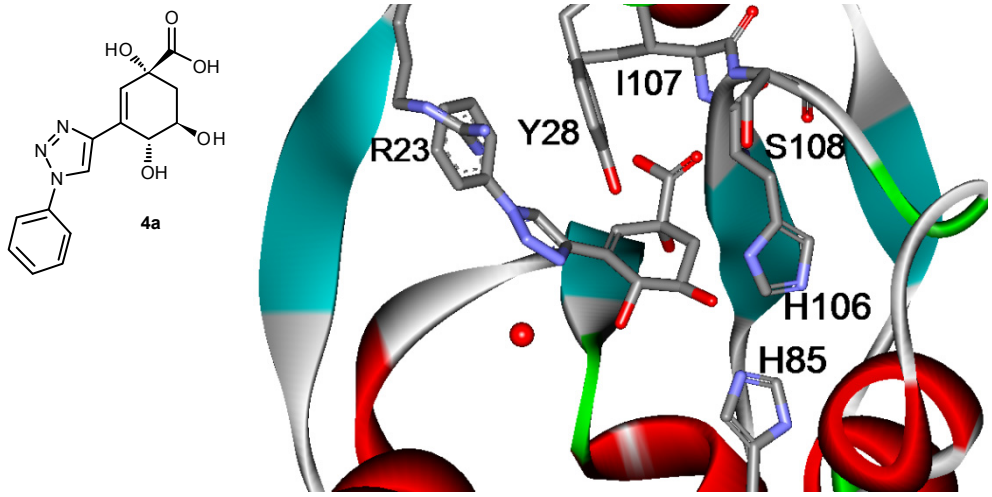


Figure 1. Docking solution of **4a** with *S. coelicolor* type II dehydroquinase.

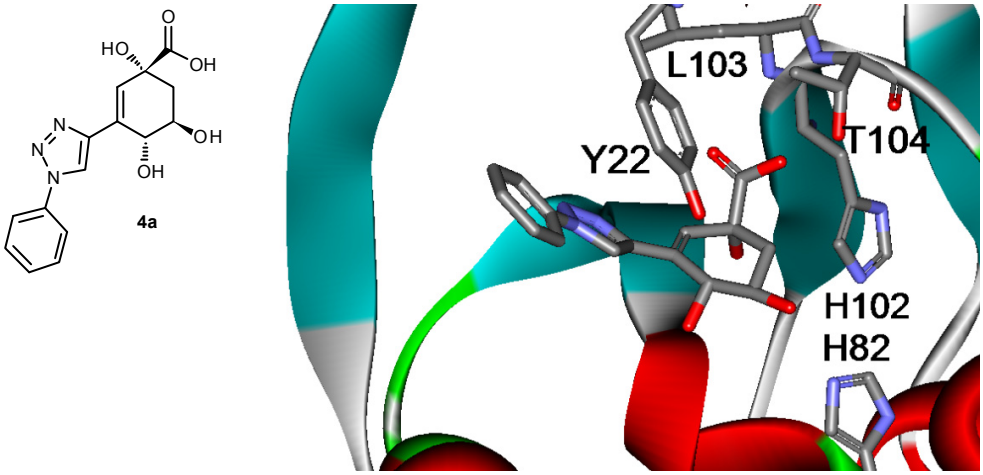


Figure 2. Docking solution of **4a** with *H. pylori* type II dehydroquinase.

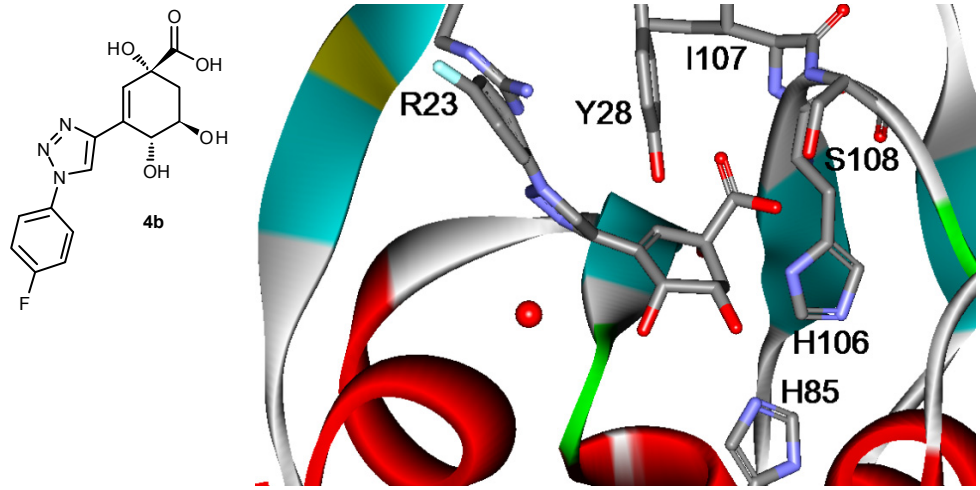


Figure 3. Docking solution of **4b** with *S. coelicolor* type II dehydroquinase.

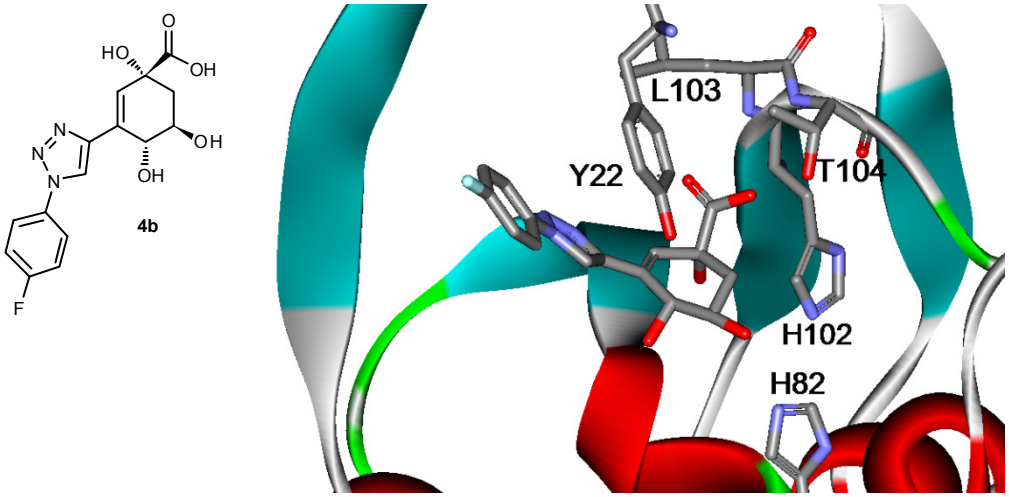


Figure 4. Docking solution of **4b** with *H. pylori* type II dehydroquinase.

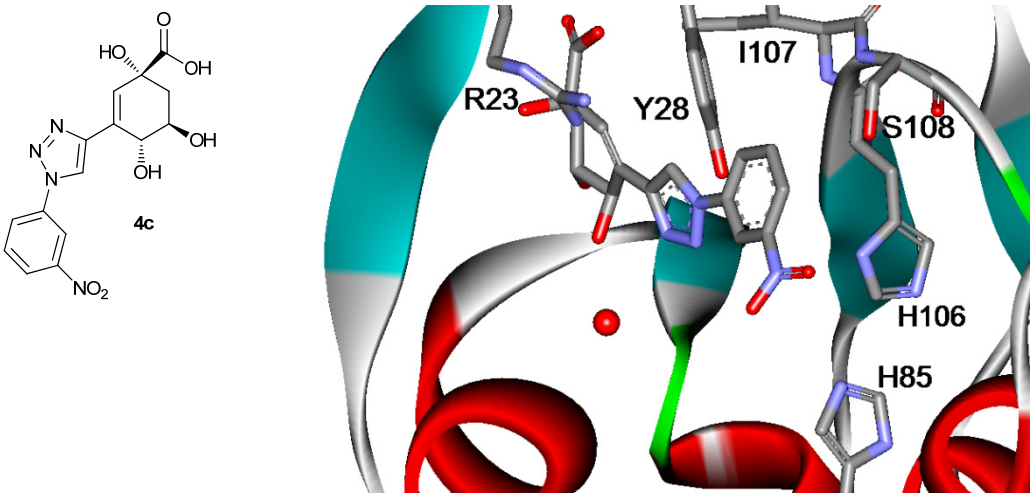


Figure 5. Docking solution of **4c** with *S. coelicolor* type II dehydroquinase.

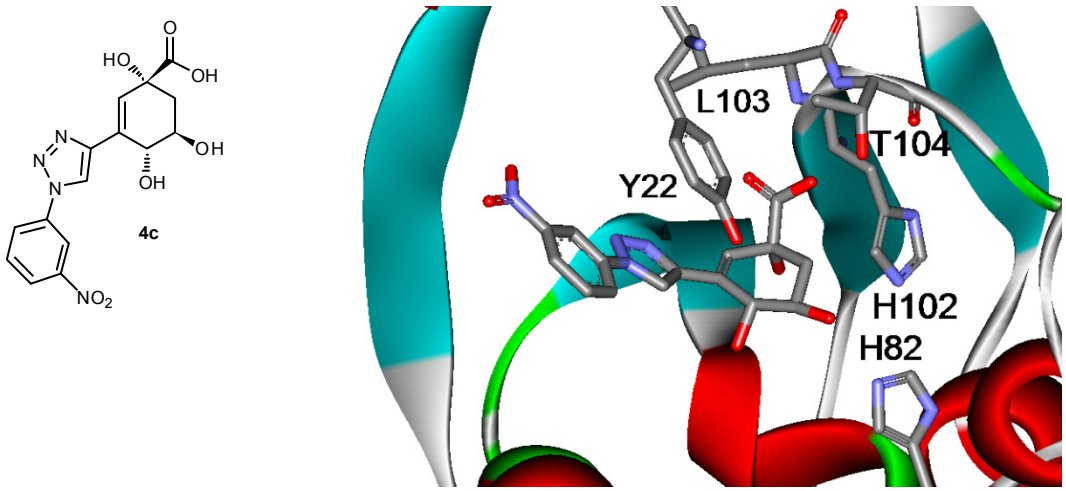


Figure 6. Docking solution of **4c** with *H. pylori* type II dehydroquinase.

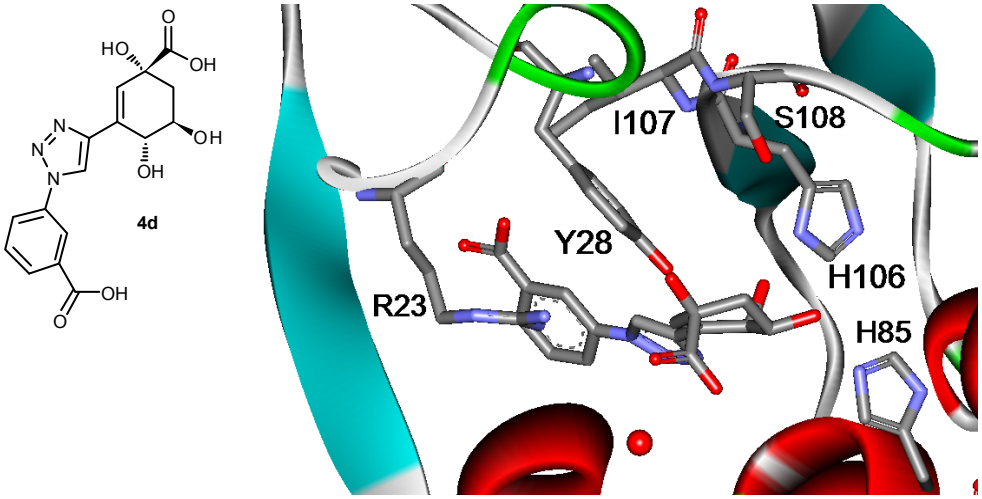


Figure 7. Docking solution of **4d** with *S. coelicolor* type II dehydroquinase.

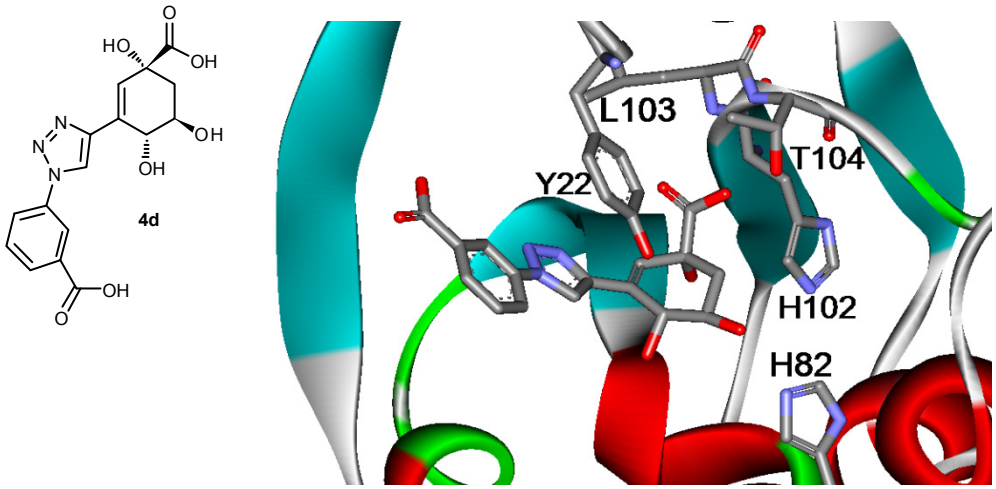


Figure 8. Docking solution of **4d** with *H. pylori* type II dehydroquinase.

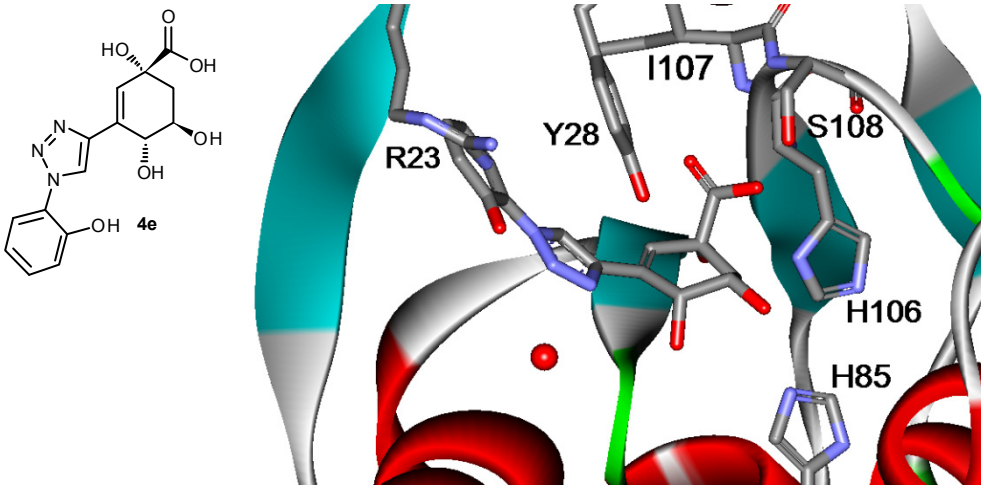


Figure 9. Docking solution of **4e** with *S. coelicolor* type II dehydroquinase.

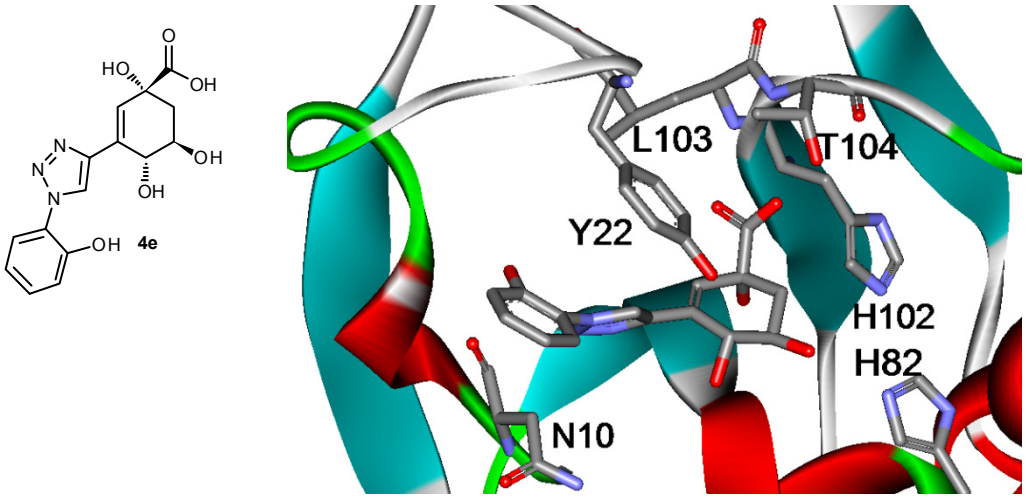


Figure 10. Docking solution of **4e** with *H. pylori* type II dehydroquinase.

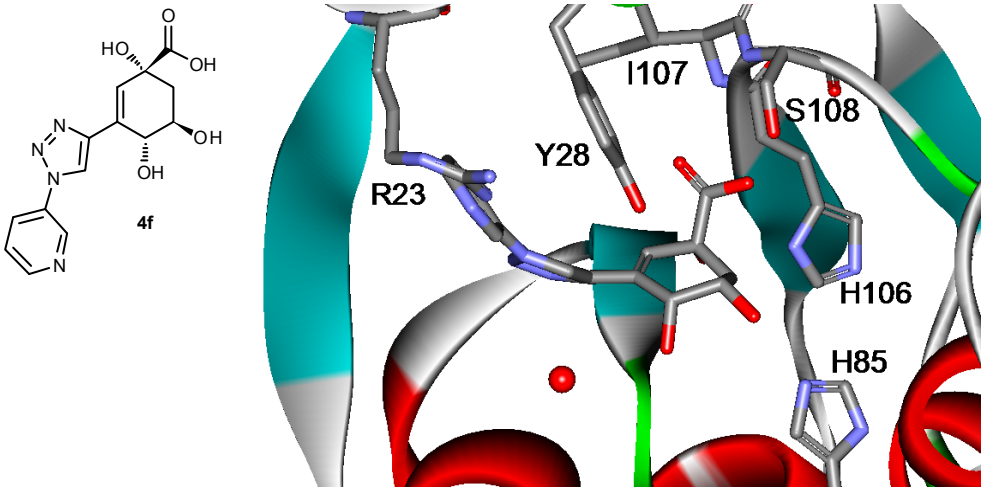


Figure 11. Docking solution of **4f** with *S. coelicolor* type II dehydroquinase.

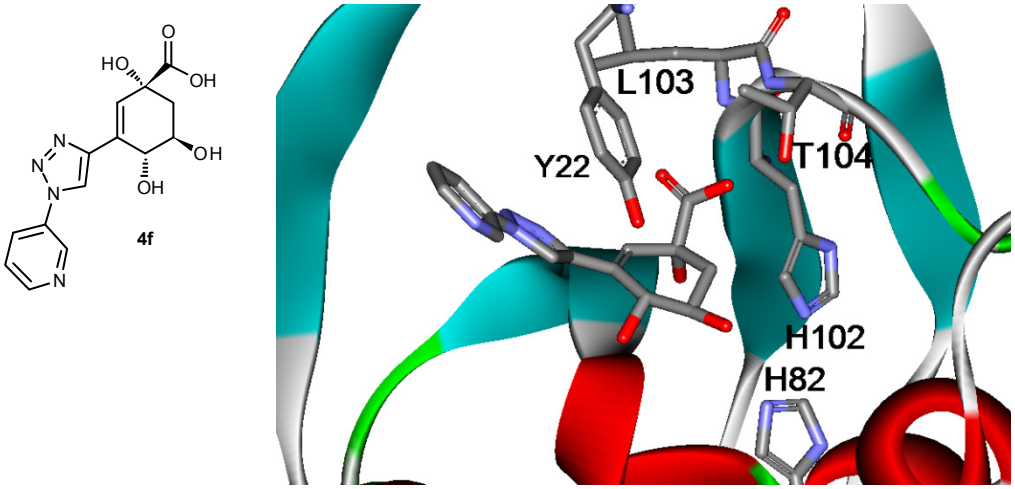


Figure 12. Docking solution of **4f** with *H. pylori* type II dehydroquinase.

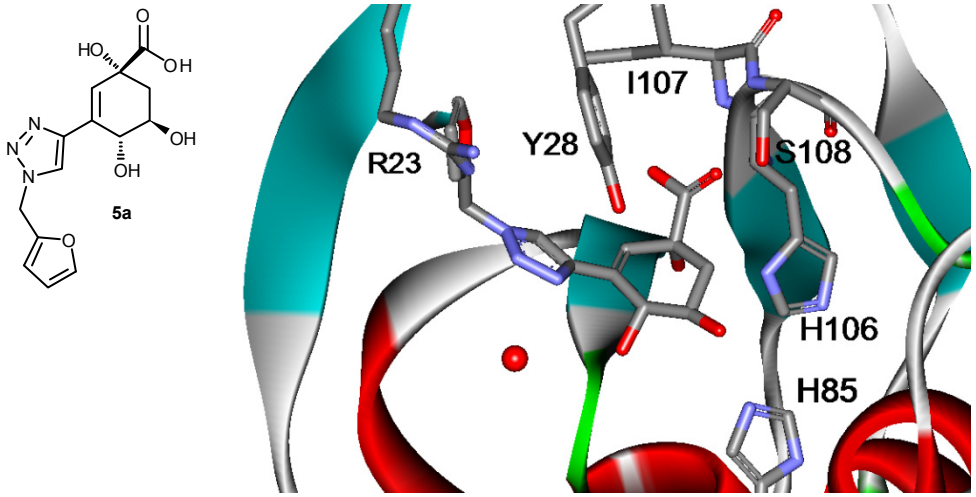


Figure 13. Docking solution of **5a** with *S. coelicolor* type II dehydroquinase.

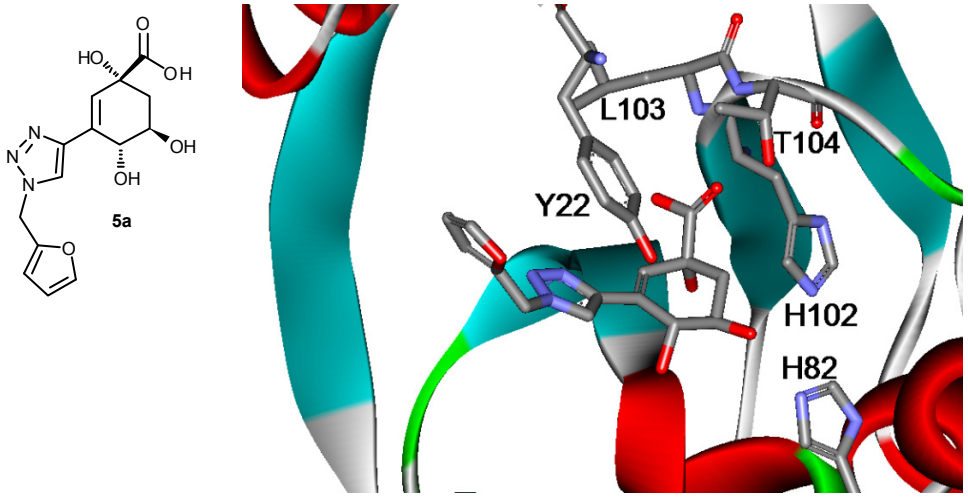


Figure 14. Docking solution of **5a** with *H. pylori* type II dehydroquinase.

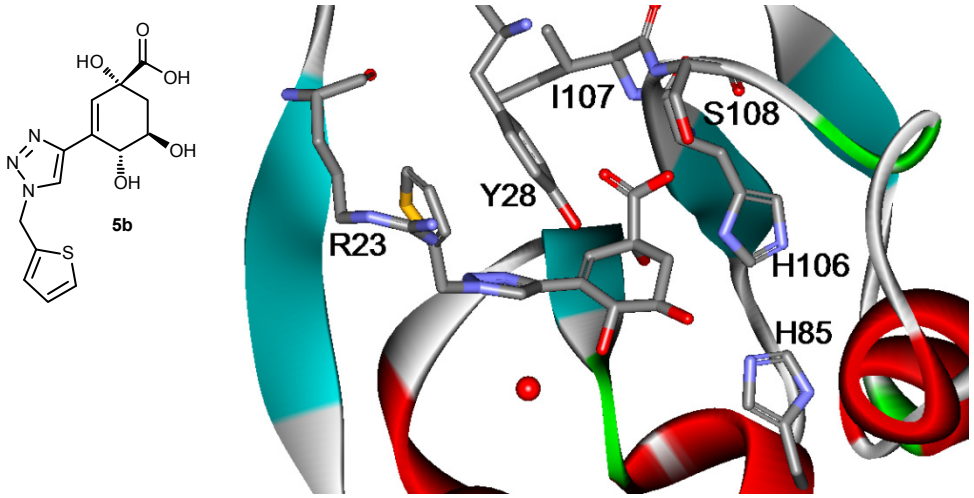


Figure 15. Docking solution of **5b** with *S. coelicolor* type II dehydroquinase.

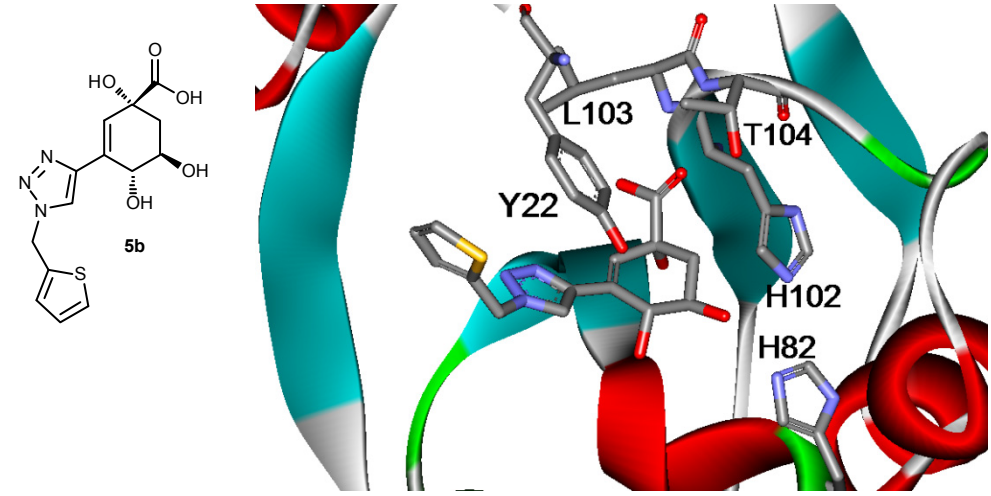
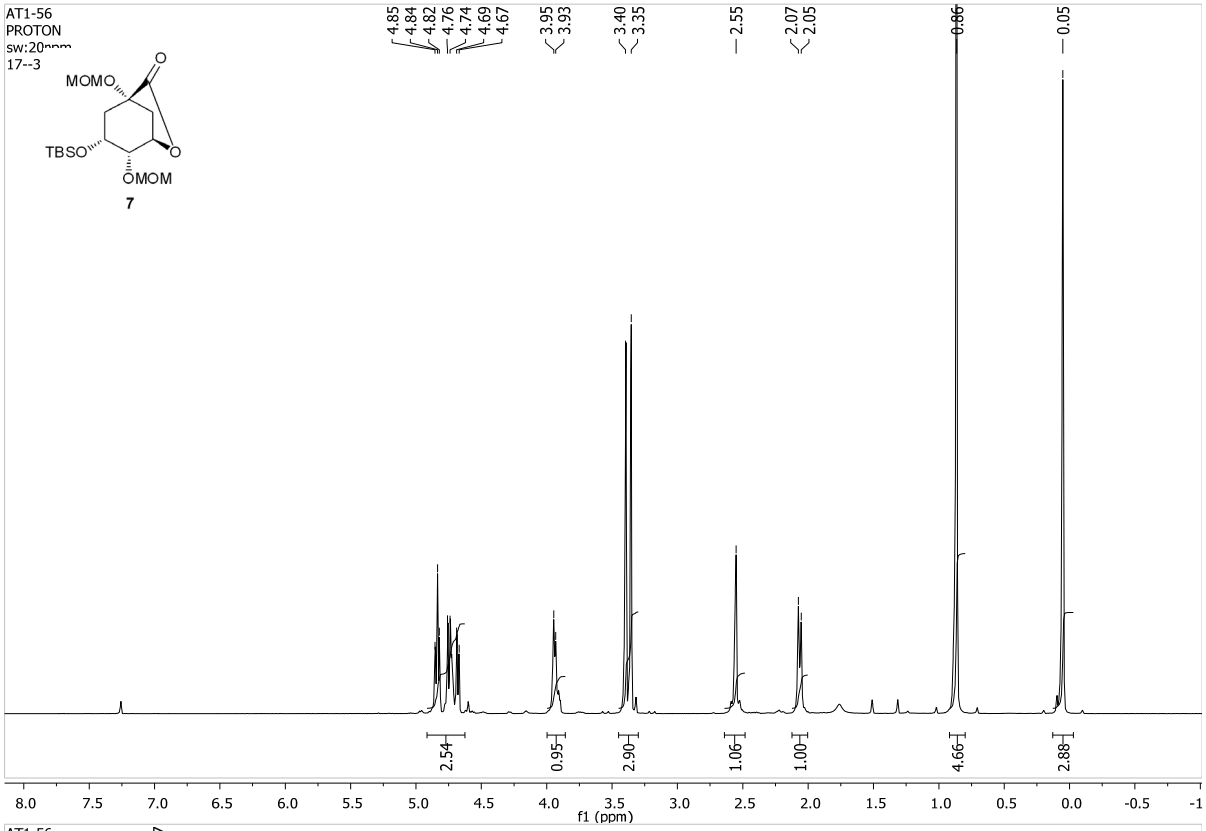
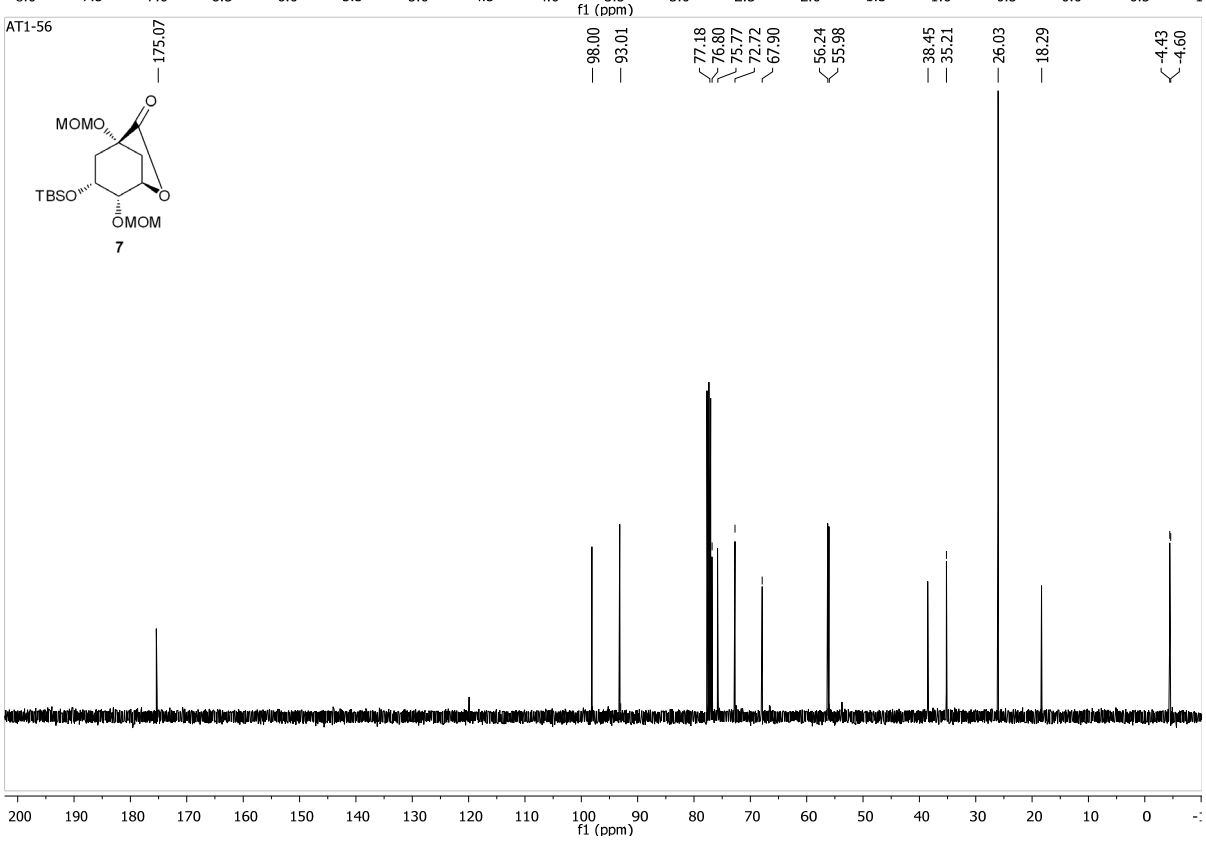
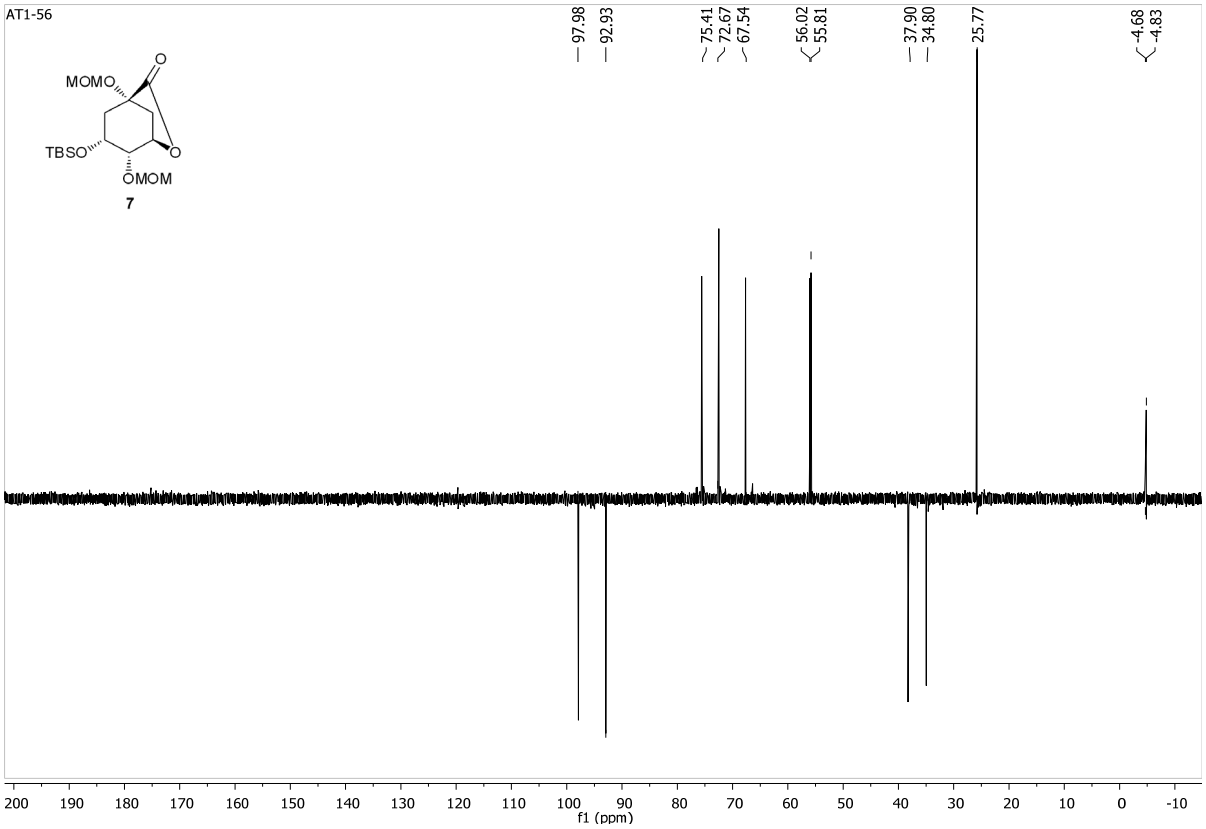


Figure 16. Docking solution of **5b** with *H. pylori* type II dehydroquinase.

^{1}H, ^{13}C NMR and DEPT-135 spectra of 7-11 and 3






AT1-56
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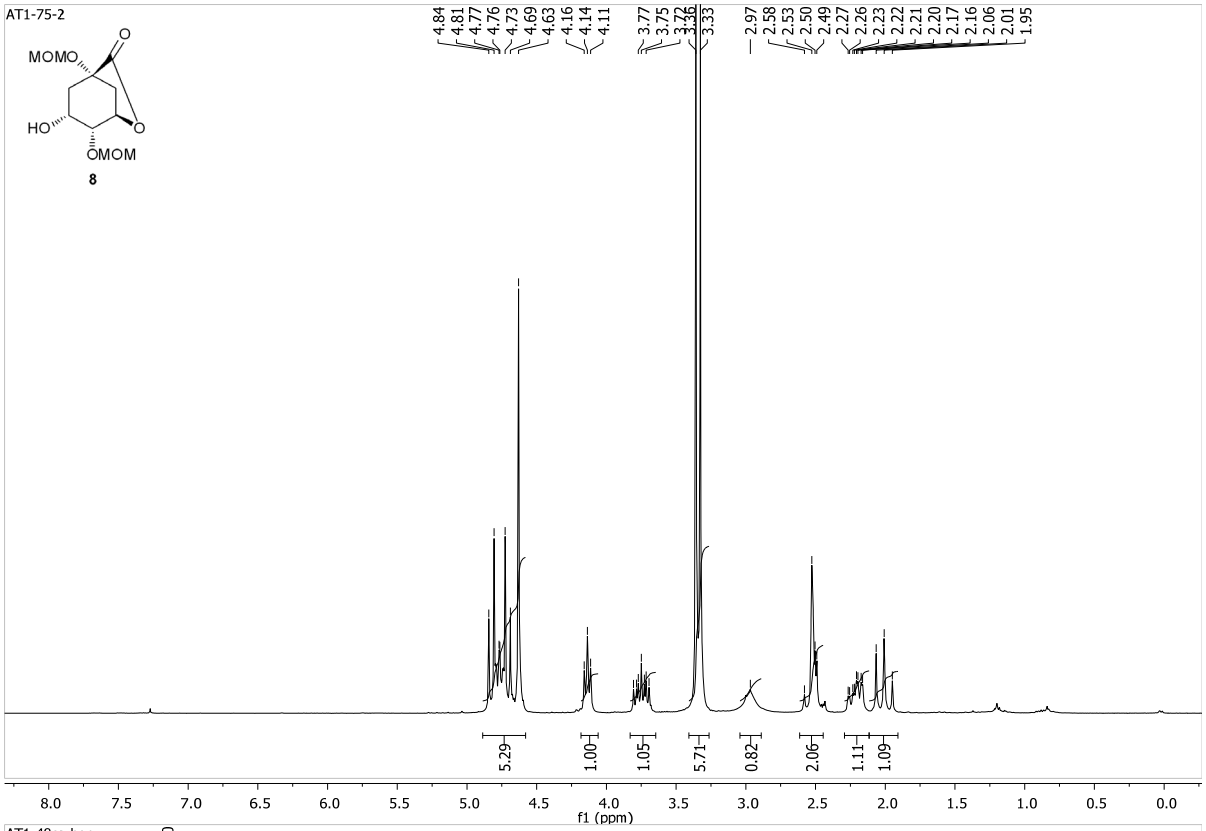
75.41
72.67
67.54
56.02
55.81
37.90
34.80
25.77
-4.68
-4.83
MOMO
TBSO
OMOM
O
O
7
200
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
0
-10
f1 (ppm)


AT1-75-2
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HO
OMOM
8
4.84
4.81
4.77
4.76
4.73
4.69
4.63
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4.14
4.11
3.77
3.75
3.72
3.36
3.33
2.97
2.58
2.53
2.50
2.49
2.27
2.26
2.23
2.22
2.21
2.20
2.17
2.16
2.06
2.01
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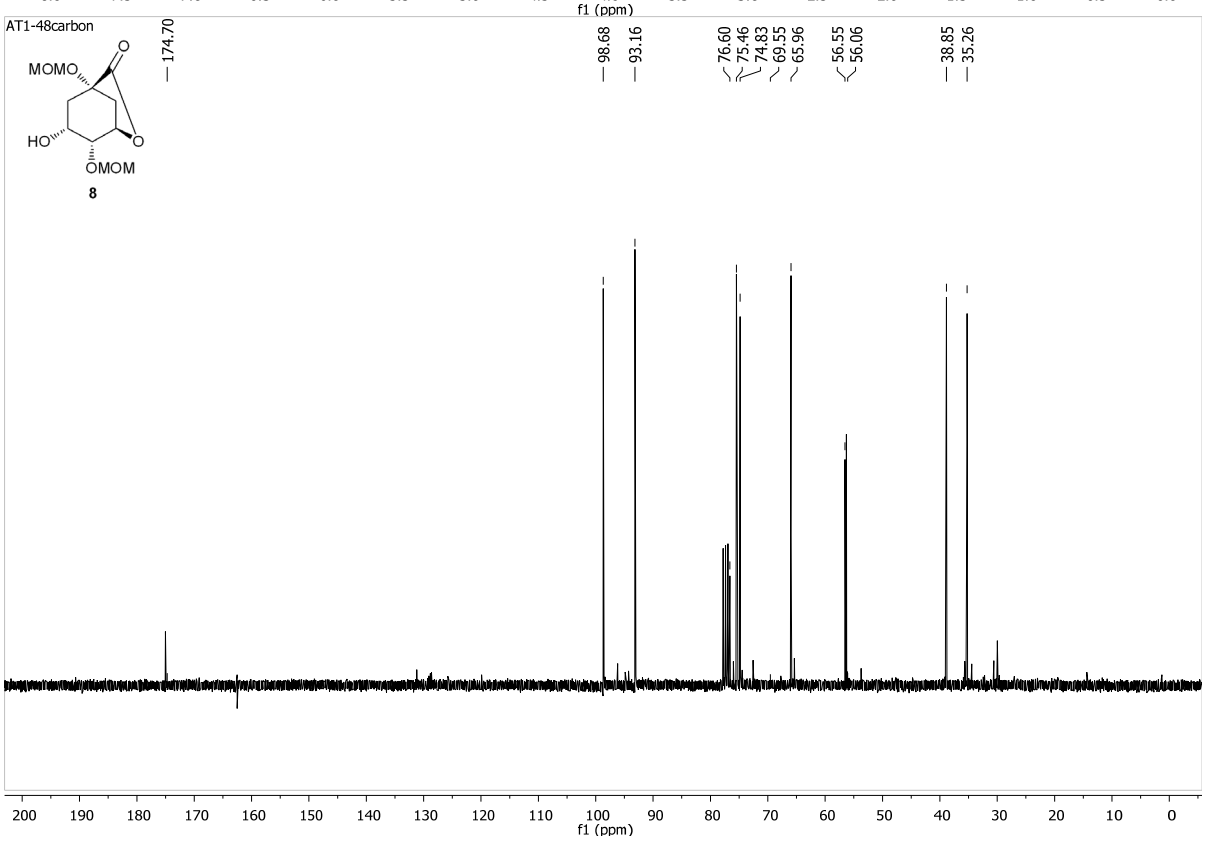
5.29
1.00
1.05
5.71
0.82
2.06
1.11
1.09
f1 (ppm)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69.55
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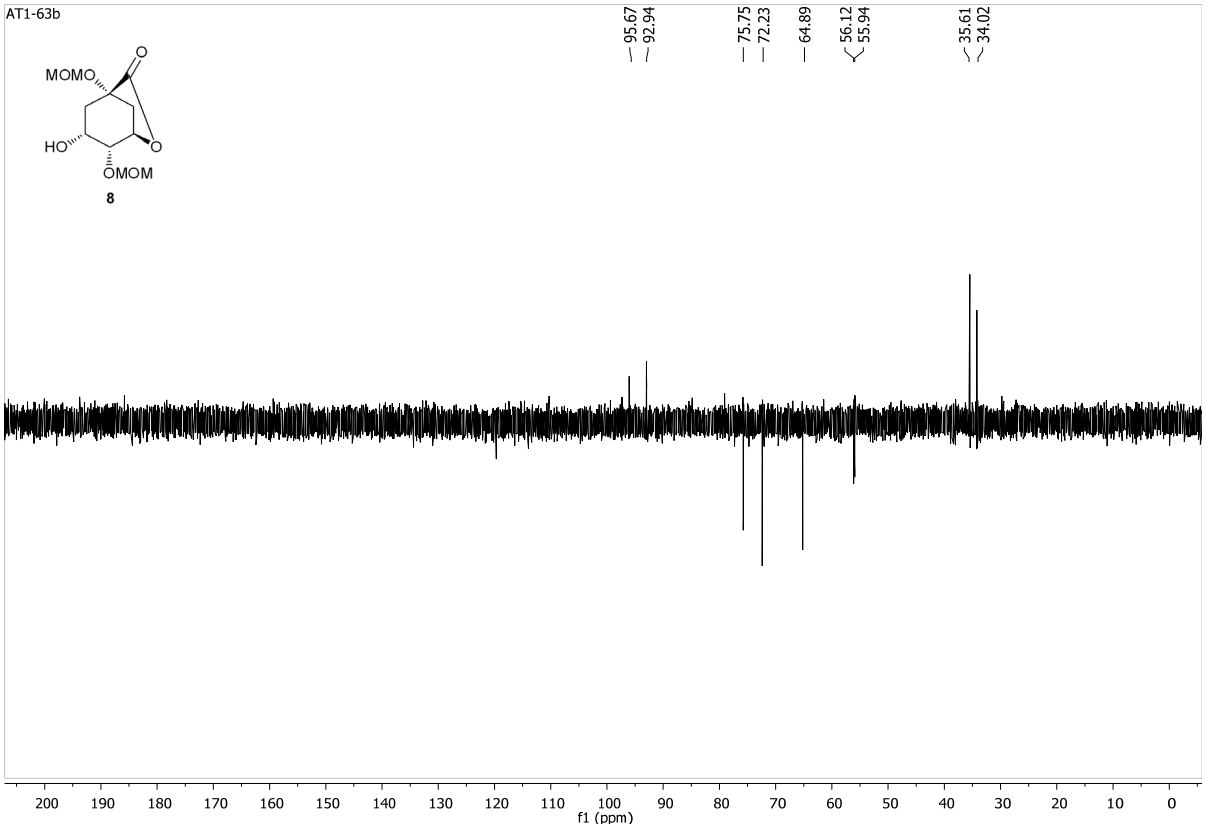
56.55
56.06
38.85
35.26
f1 (ppm)


AT1-63b
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75.75
72.23
64.89
56.12
55.94
35.61
34.02
MOMO
HO
OMOM
O
O
8
200
190
180
170
160
150
140
130
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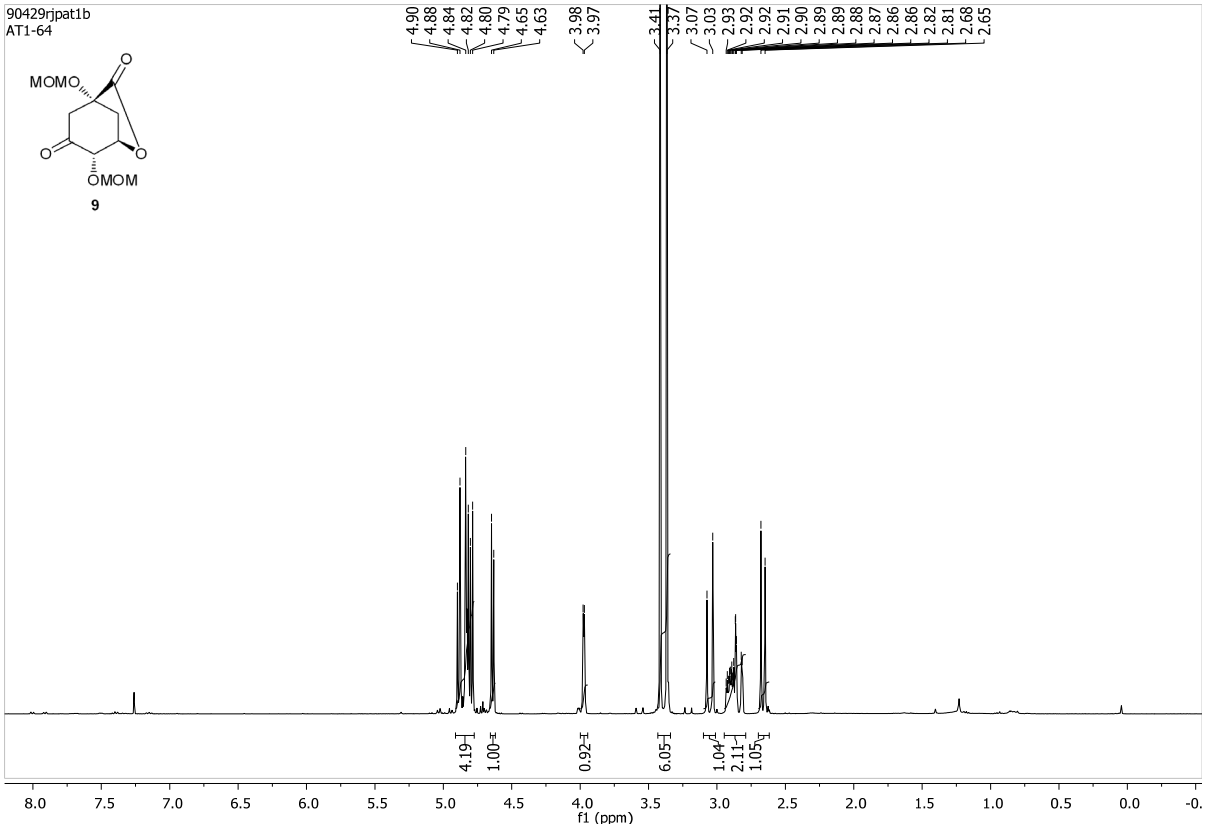
110
100
90
80
70
60
50
40
30
20
10
0
f1 (ppm)


90429rjpat1b
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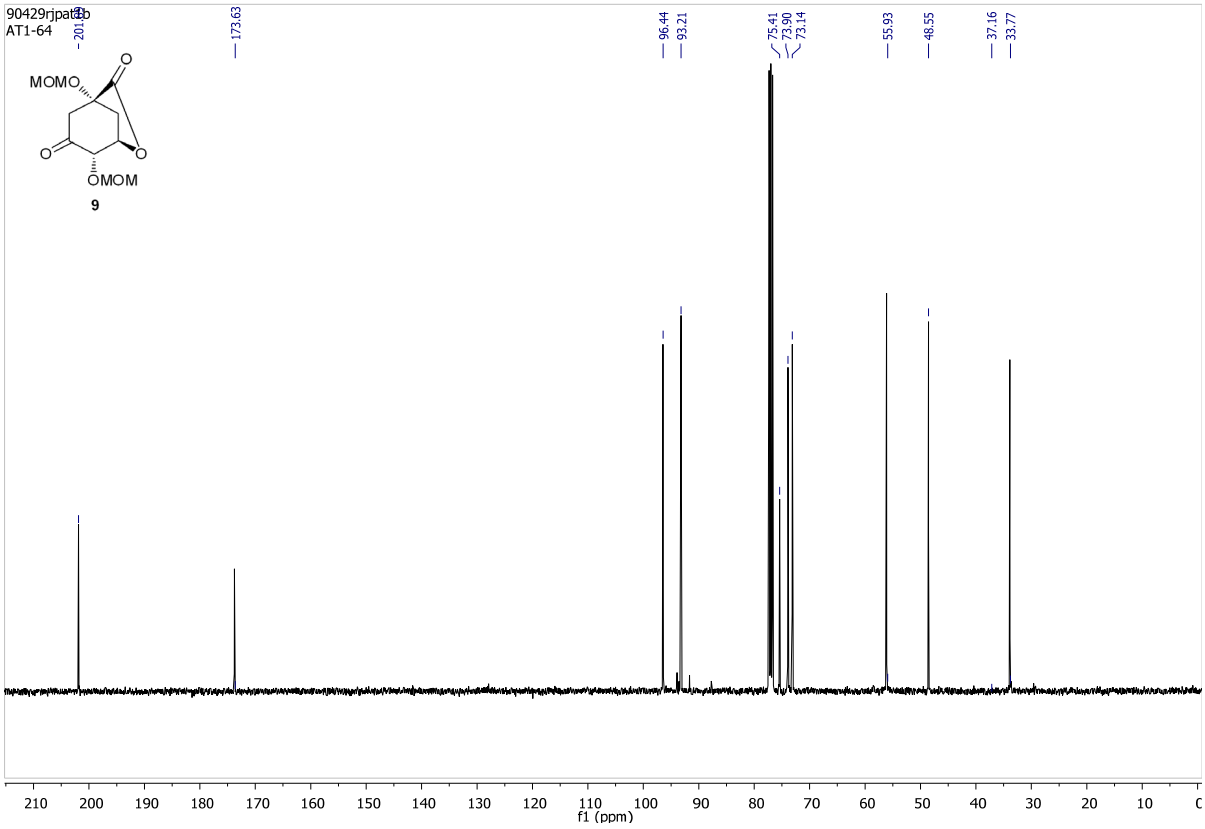
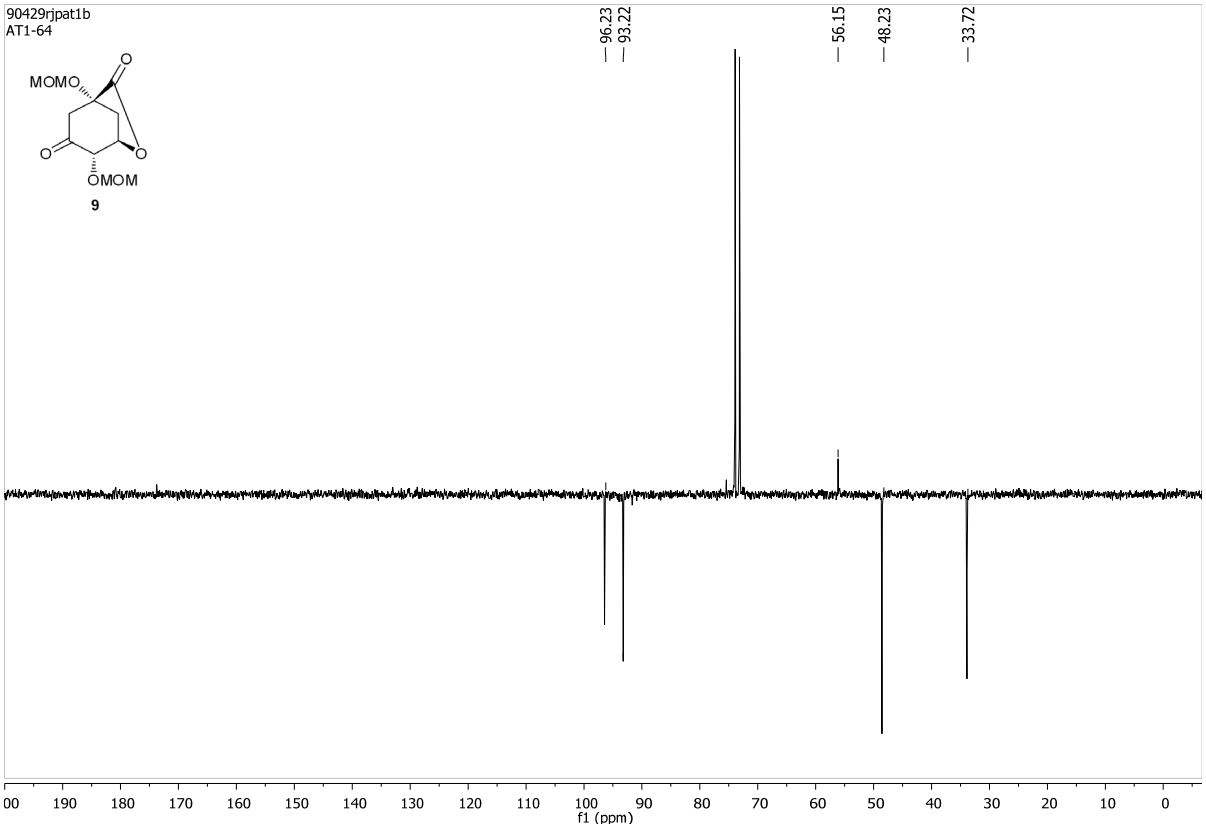
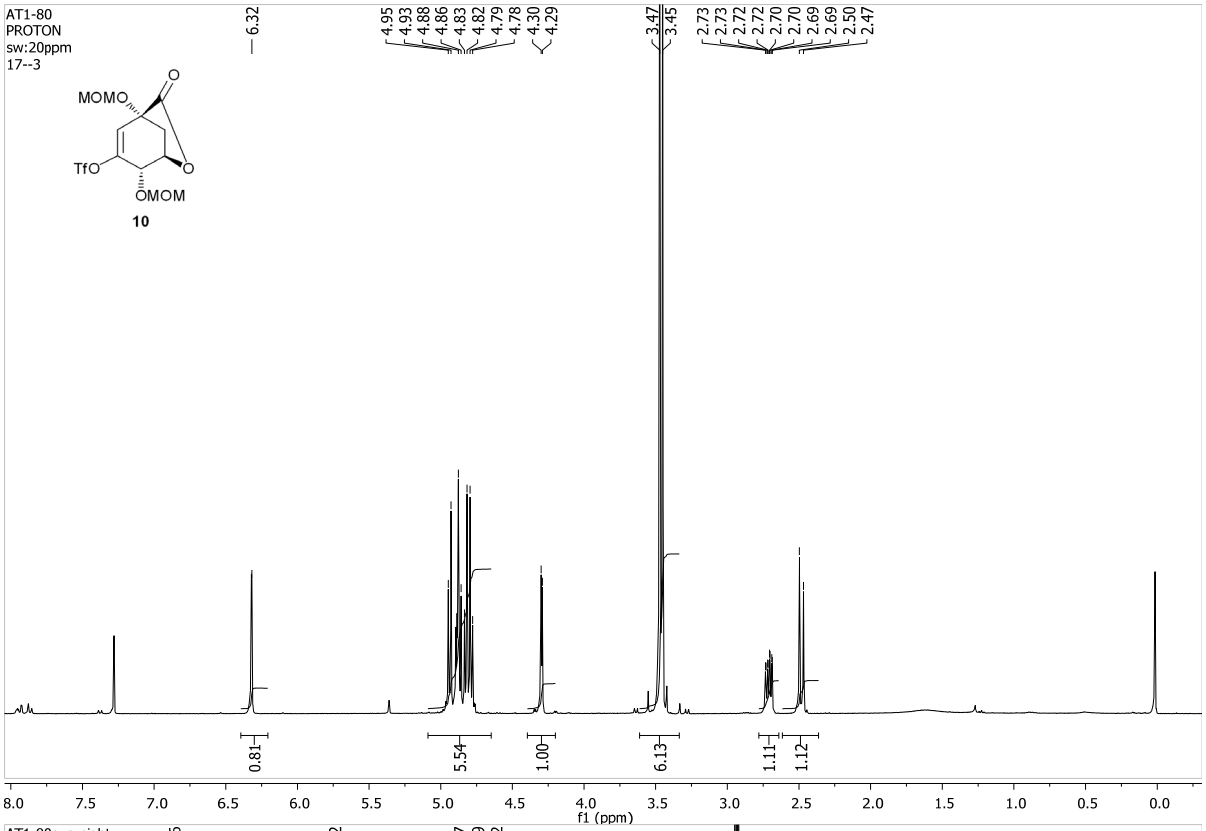
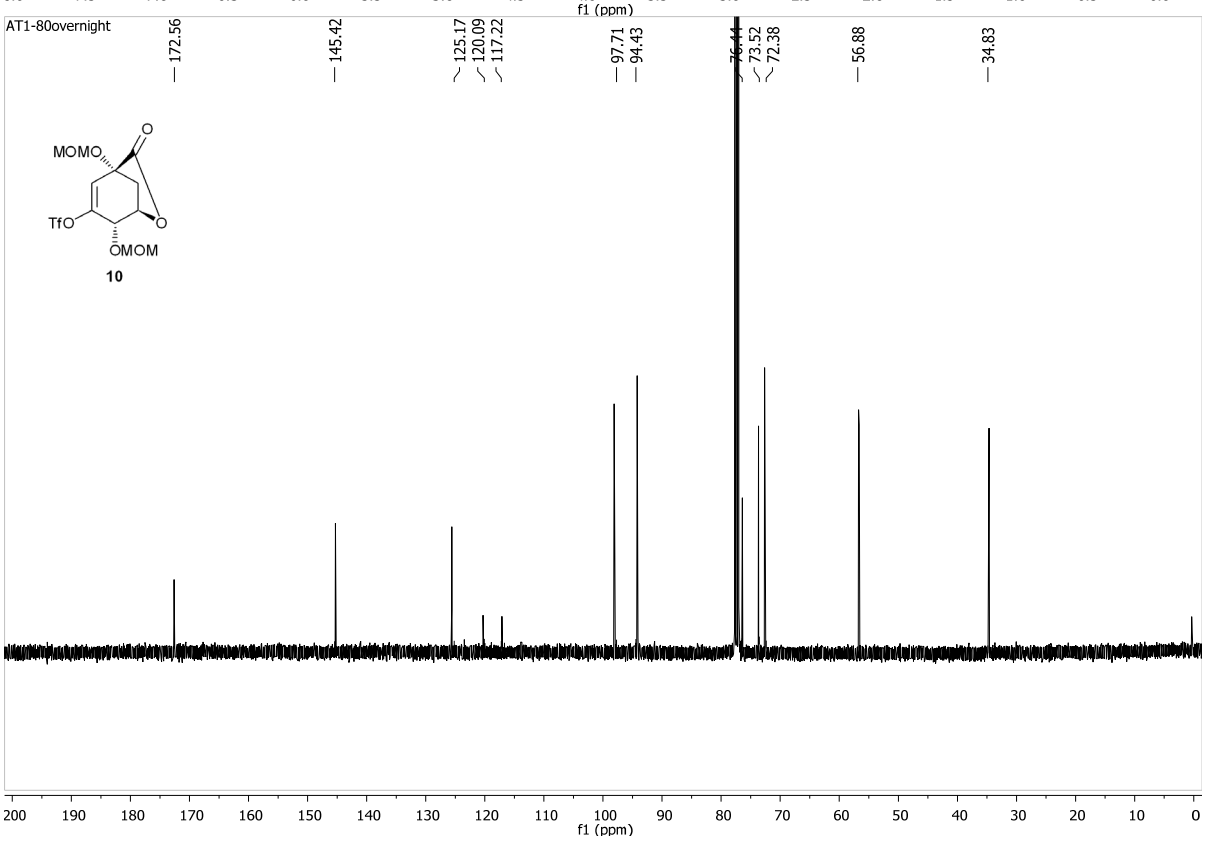
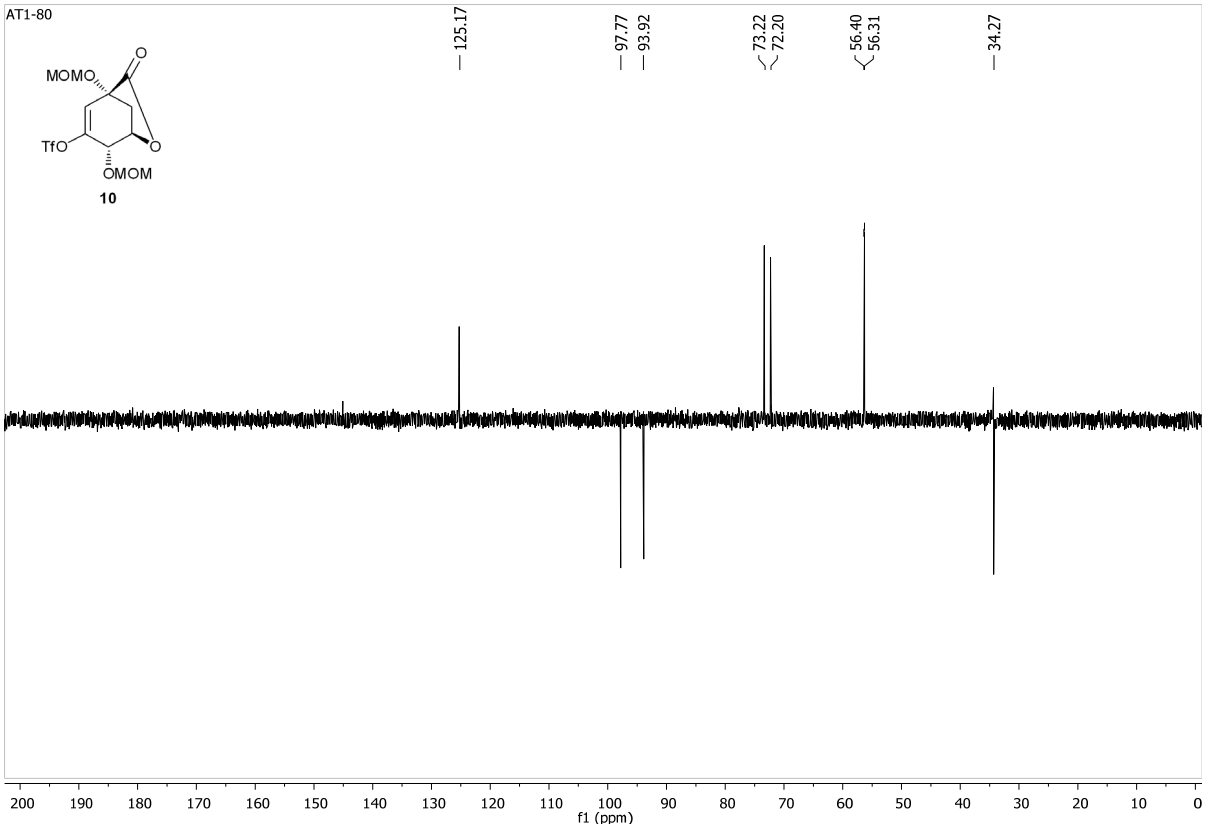
MOMO
OMOM
9
4.90
4.88
4.84
4.82
4.80
4.79
4.65
4.63
3.98
3.97
3.41
3.37
3.07
3.03
2.93
2.92
2.92
2.91
2.90
2.89
2.89
2.88
2.87
2.86
2.86
2.82
2.81
2.68
2.65
4.19
1.00
0.92
6.05
1.04
2.11
1.05
f1 (ppm)

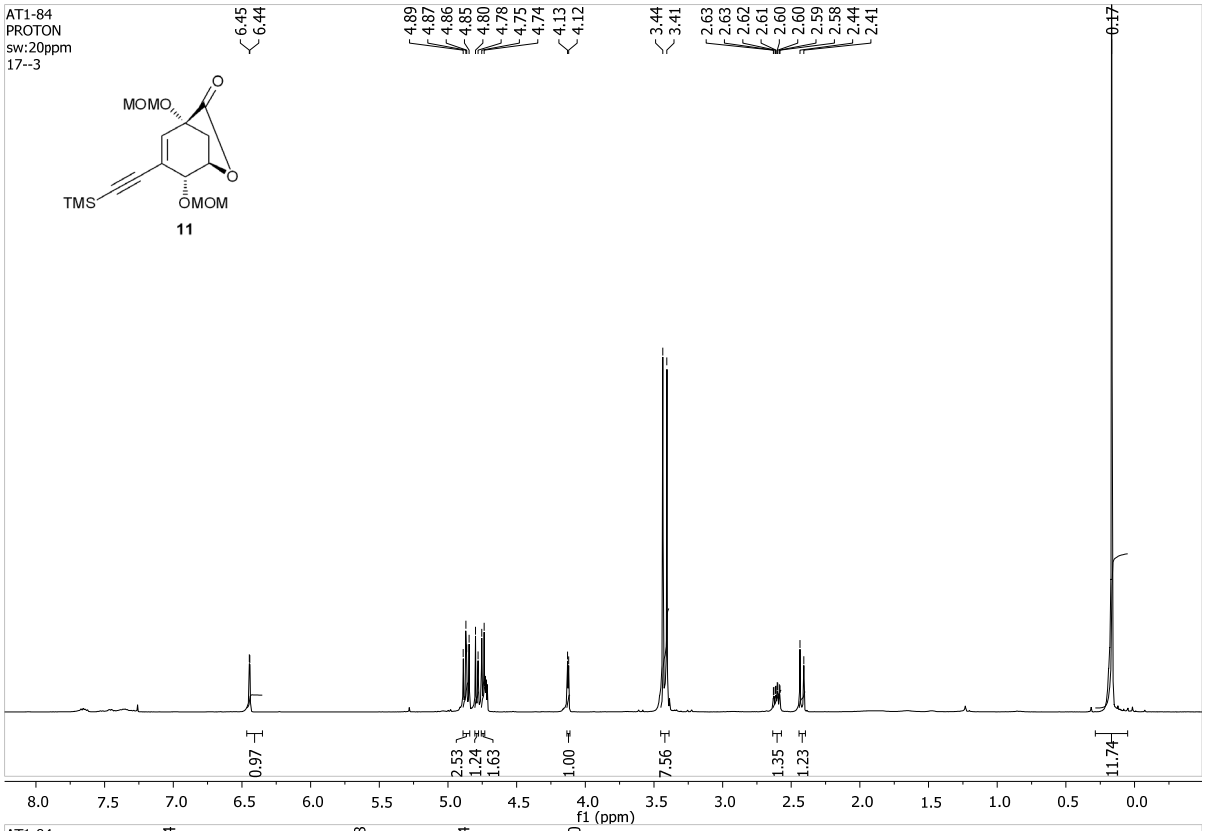

90429rjpat3b
AT1-64
MOMO
OMOM
9
201.48
173.63
96.44
93.21
75.41
73.90
73.14
55.93
48.55
37.16
33.77
f1 (ppm)

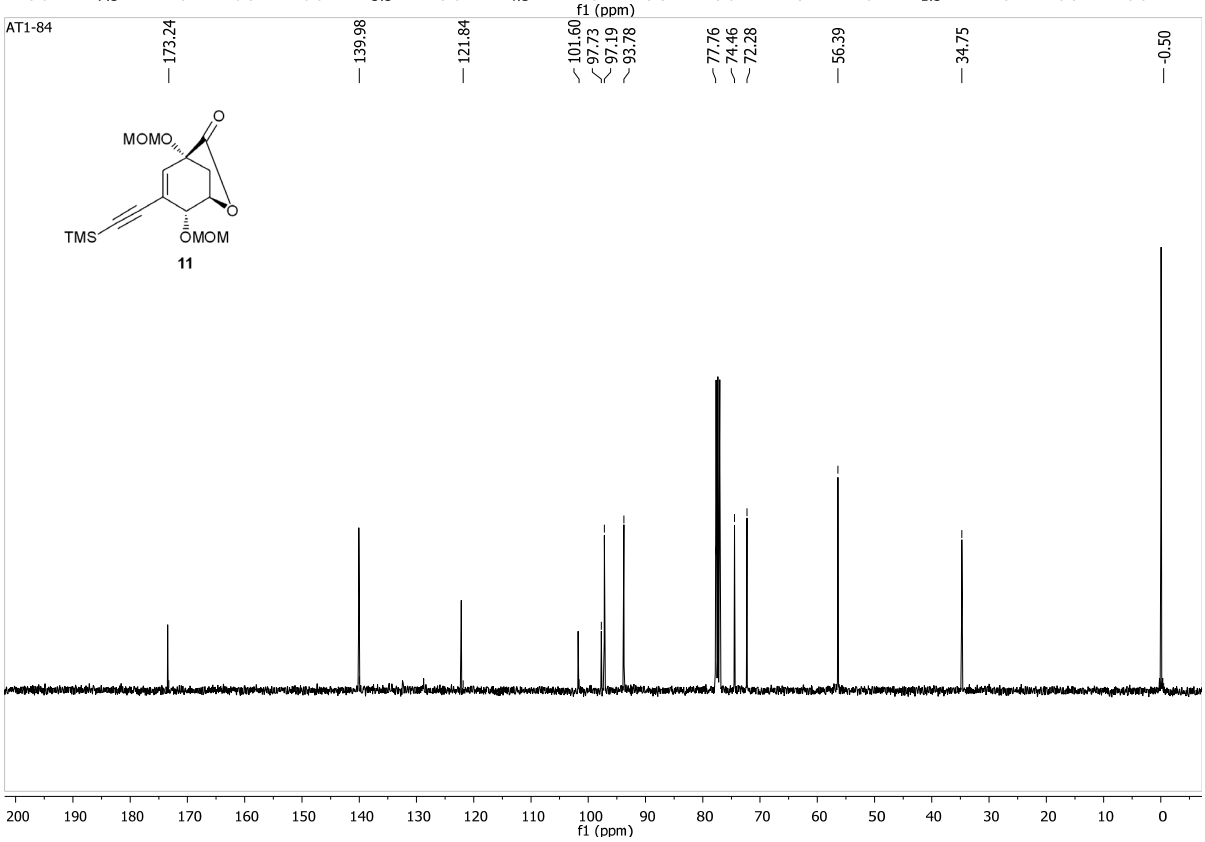

90429rjpat1b
AT1-64
96.23
93.22
56.15
48.23
33.72
MOMO
OMOM
9
f1 (ppm)

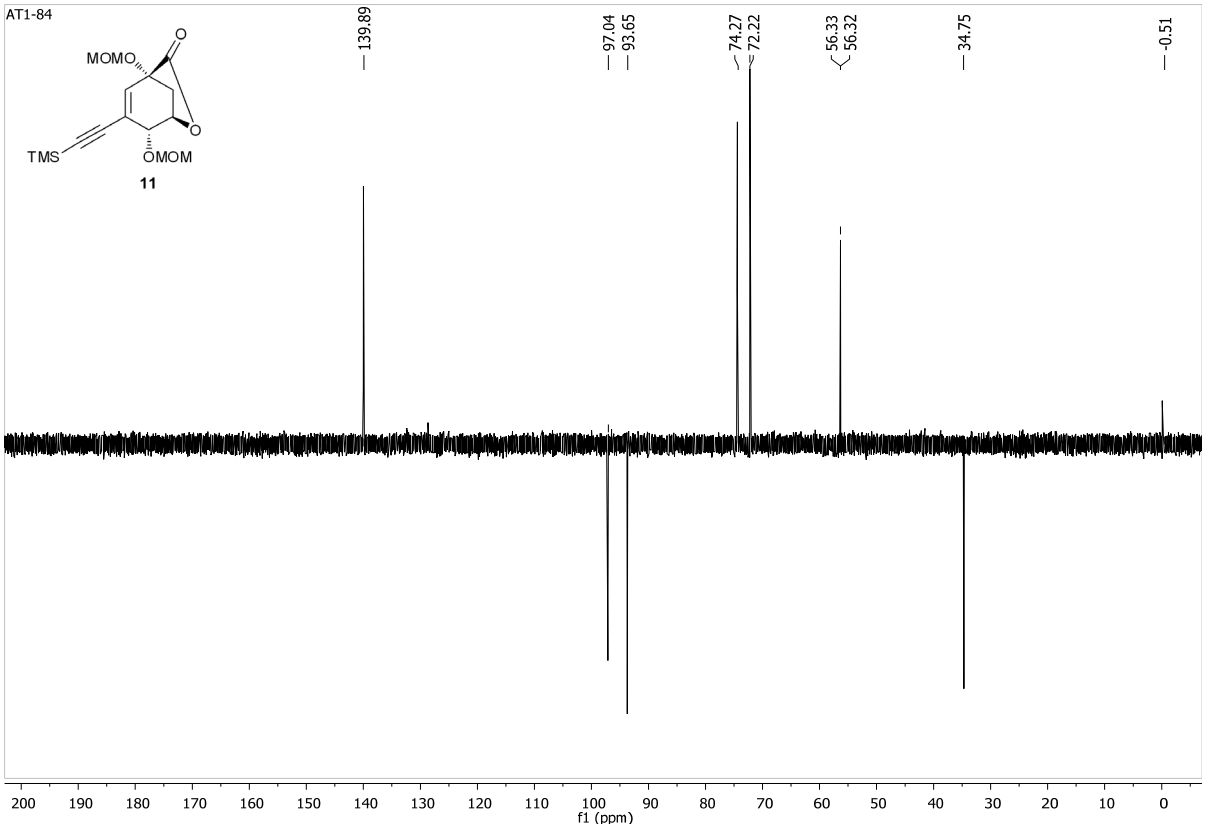

AT1-80
PROTON
sw:20ppm
17--3
6.32
4.95
4.93
4.88
4.86
4.83
4.82
4.79
4.78
4.30
4.29
3.47
3.45
2.73
2.73
2.72
2.72
2.70
2.70
2.69
2.69
2.50
2.47
MOMO
TfO
OMOM
O
10
0.81
5.54
1.00
6.13
1.11
1.12
f1 (ppm)

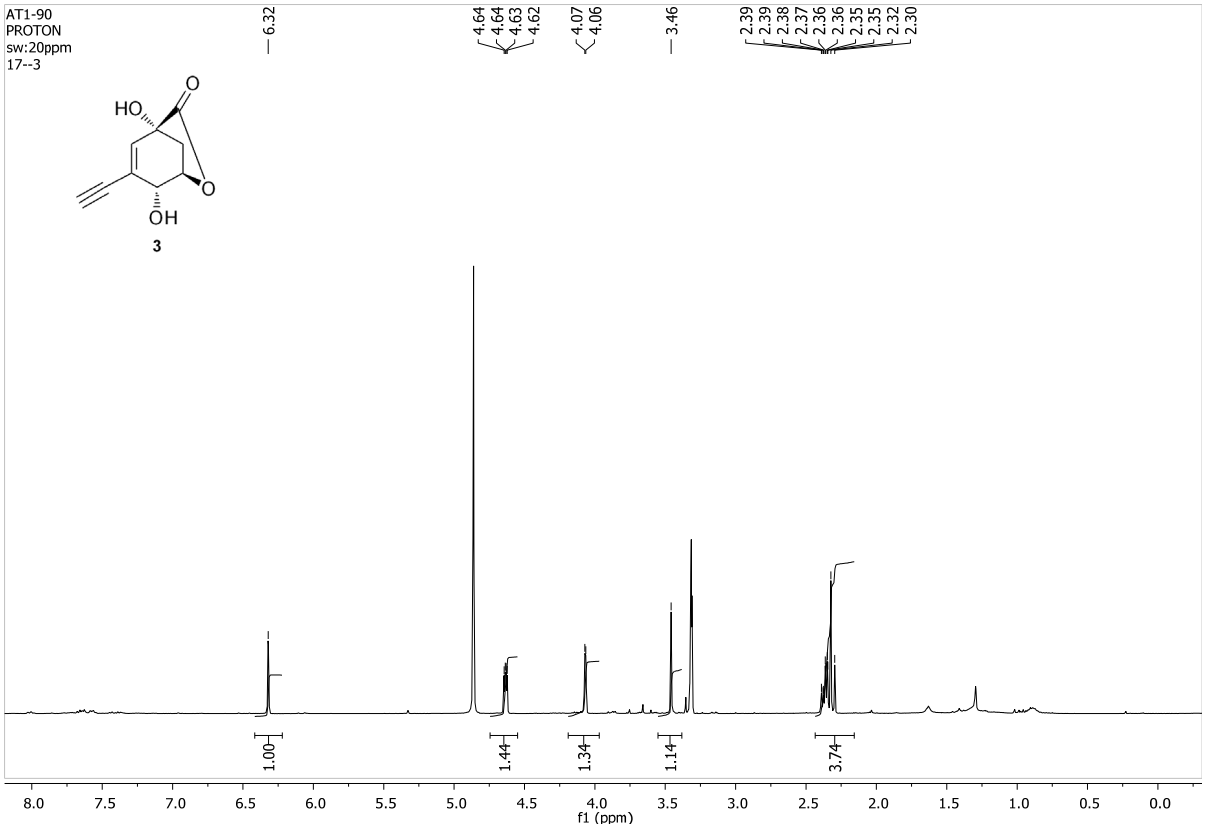

AT1-80overnight
172.56
145.42
125.17
120.09
117.22
97.71
94.43
76.44
73.52
72.38
56.88
34.83
MOMO
TfO
OMOM
O
10
f1 (ppm)


AT1-80
MOMO
TfO
OMOM
O
O
10
125.17
97.77
93.92
73.22
72.20
56.40
56.31
34.27
200
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
0
f1 (ppm)

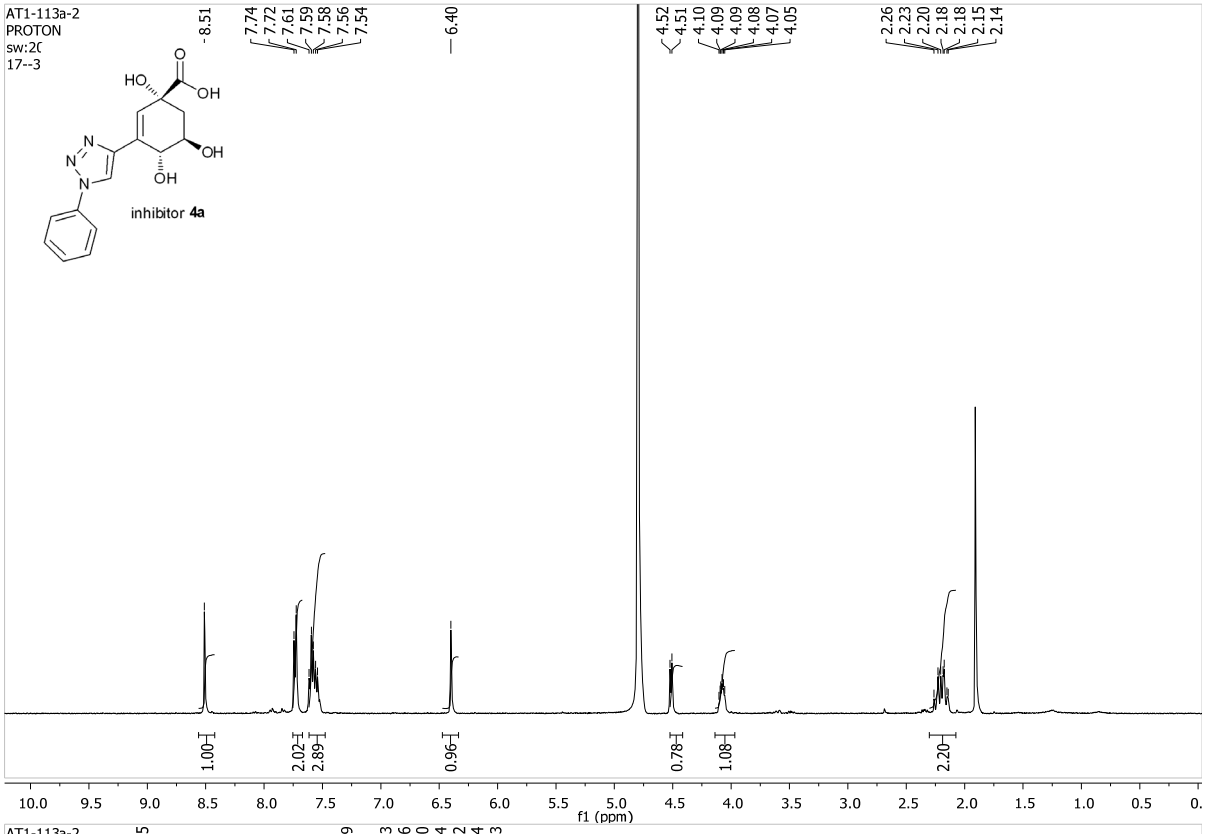

AT1-84
PROTON
sw:20ppm
17--3
6.45
6.44
4.89
4.87
4.86
4.85
4.80
4.78
4.75
4.74
4.13
4.12
3.44
3.41
2.63
2.63
2.62
2.61
2.60
2.60
2.59
2.58
2.44
2.41
0.17
MOMO
TMS
OMOM
11
0.97
2.53
1.24
1.63
1.00
7.56
1.35
1.23
11.74
f1 (ppm)

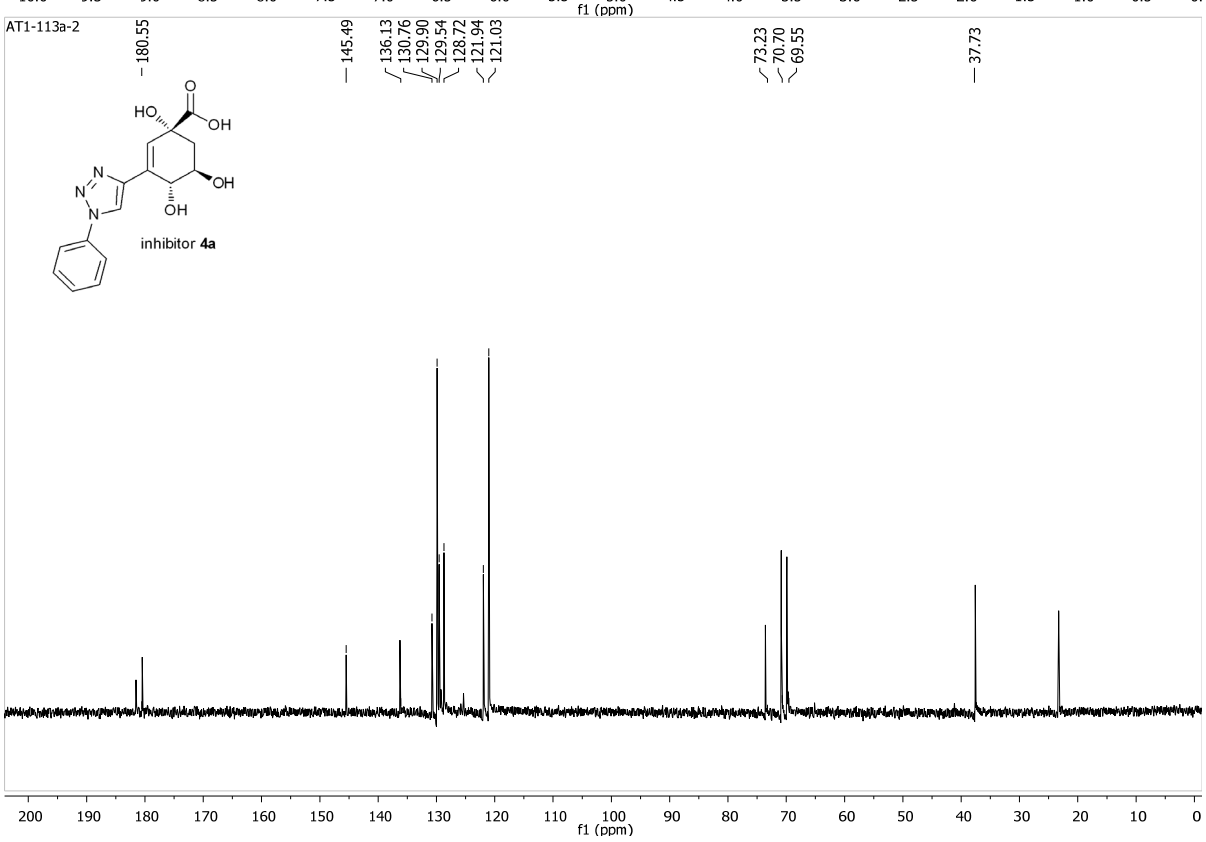

AT1-84
173.24
139.98
121.84
101.60
97.73
97.19
93.78
77.76
74.46
72.28
56.39
34.75
-0.50
MOMO
TMS
OMOM
11
f1 (ppm)

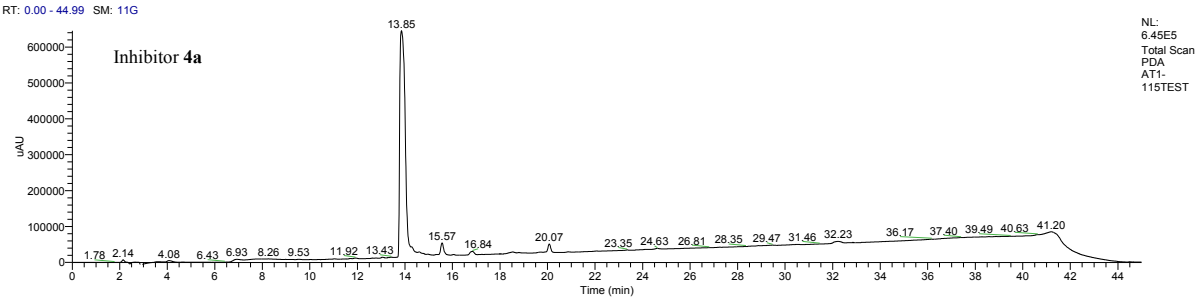

AT1-84
MOMO
TMS
OMOM
11
139.89
97.04
93.65
74.27
72.22
56.33
56.32
34.75
-0.51
200
190
180
170
160
150
140
130
120
110
100
90
80
70
60
50
40
30
20
10
0
f1 (ppm)

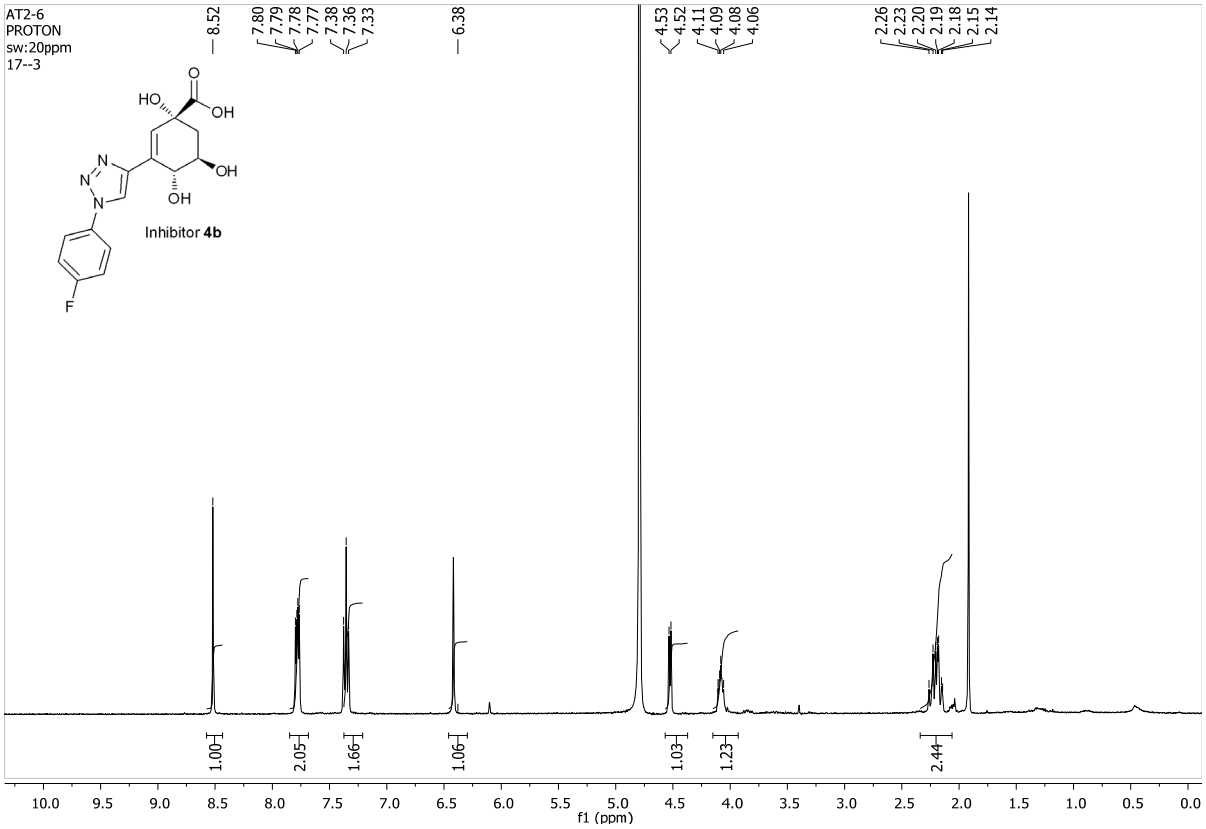

AT1-90
PROTON
sw:20ppm
17--3
6.32
4.64
4.64
4.63
4.62
4.07
4.06
3.46
2.39
2.39
2.38
2.37
2.36
2.36
2.35
2.35
2.32
2.30
HO
O
O
OH
3
1.00
1.44
1.34
1.14
3.74
8.0
7.5
7.0
6.5
6.0
5.5
5.0
4.5
4.0
3.5
3.0
2.5
2.0
1.5
1.0
0.5
0.0
f1 (ppm)

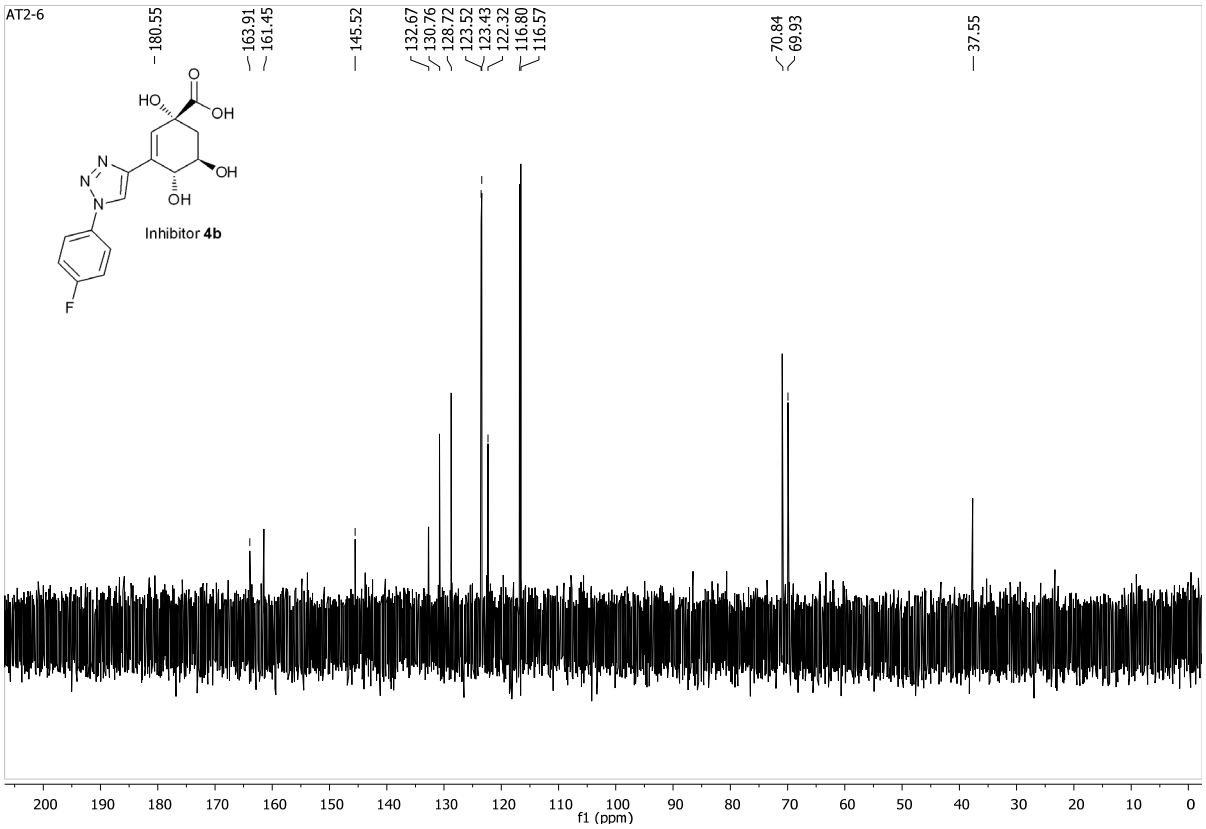
^{1}H, ^{13}C NMR and LC-MS traces of 4a-f, 5a and 5b

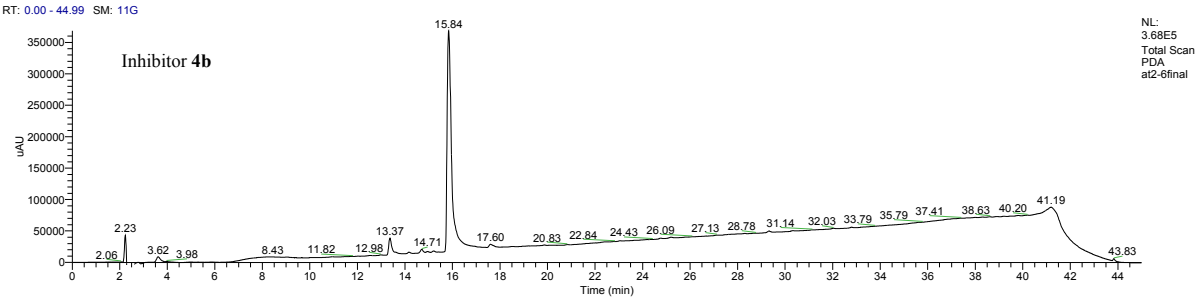


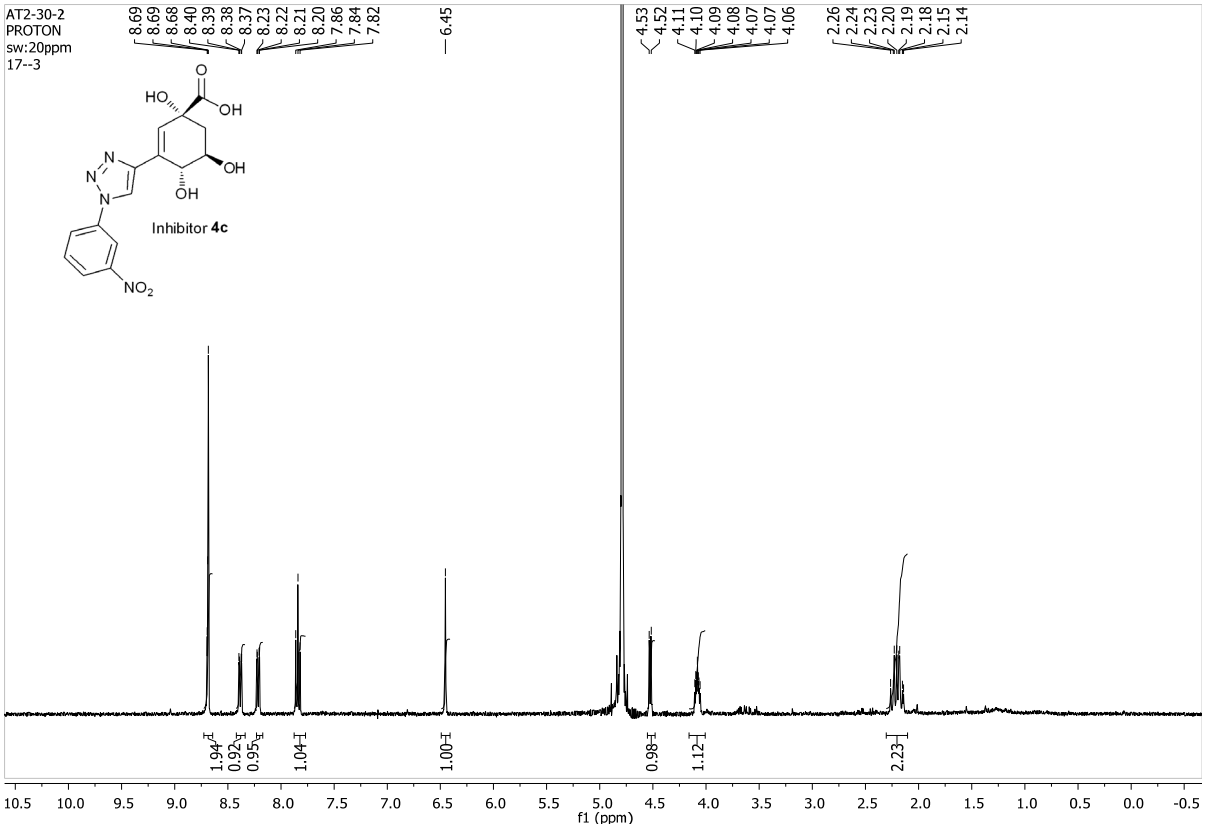


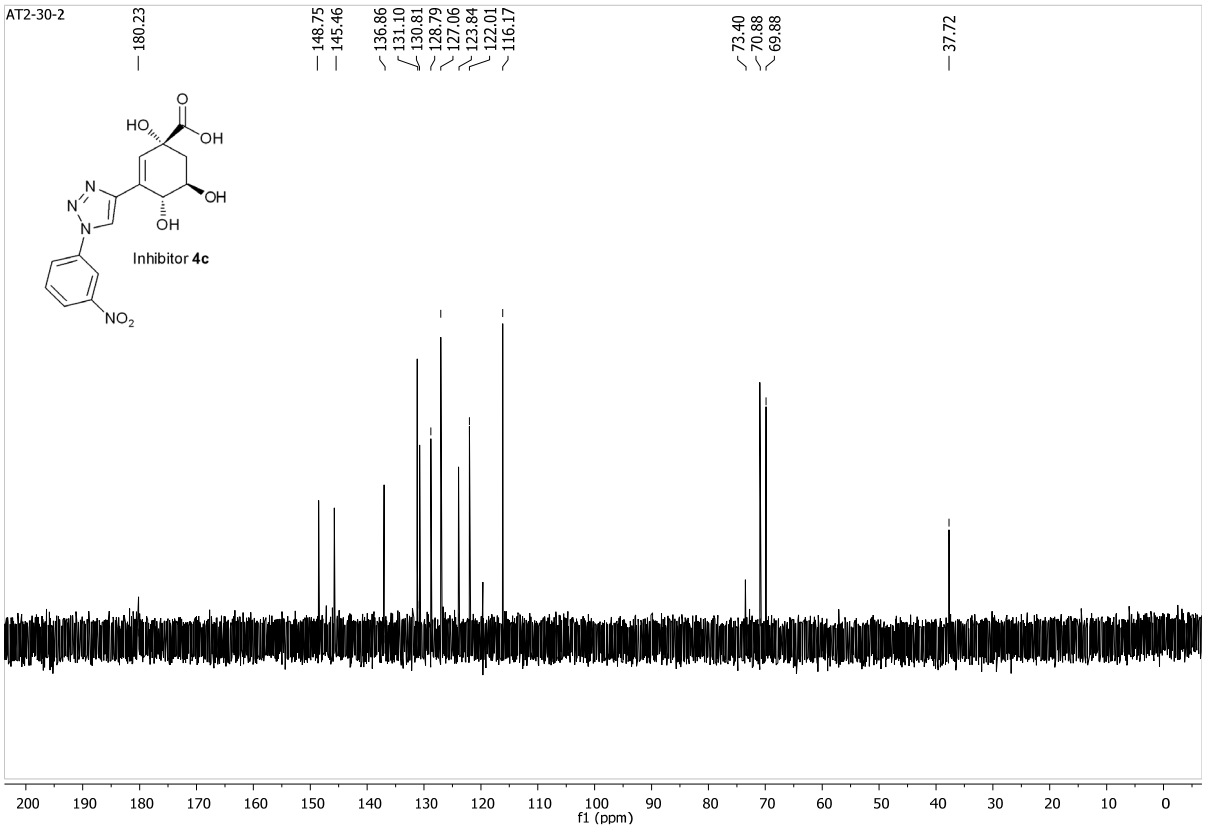

RT: 0.00 - 44.99 SM: 11G
Inhibitor 4a
NL:
6.45E5
Total Scan
PDA
AT1-
115TEST
uAU
Time (min)
13.85
1.78
2.14
4.08
6.43
6.93
8.26
9.53
11.92
13.43
15.57
16.84
20.07
23.35
24.63
26.81
28.35
29.47
31.46
32.23
36.17
37.40
39.49
40.63
41.20

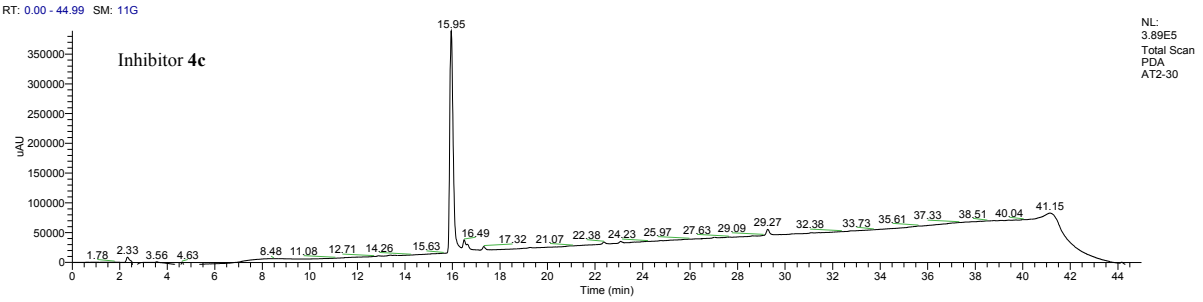

AT2-6
PROTON
sw:20ppm
17--3
8.52
7.80
7.79
7.78
7.77
7.38
7.36
7.33
6.38
4.53
4.52
4.11
4.09
4.08
4.06
2.26
2.23
2.20
2.19
2.18
2.15
2.14
Inhibitor 4b
1.00
2.05
1.66
1.06
1.03
1.23
2.44
f1 (ppm)

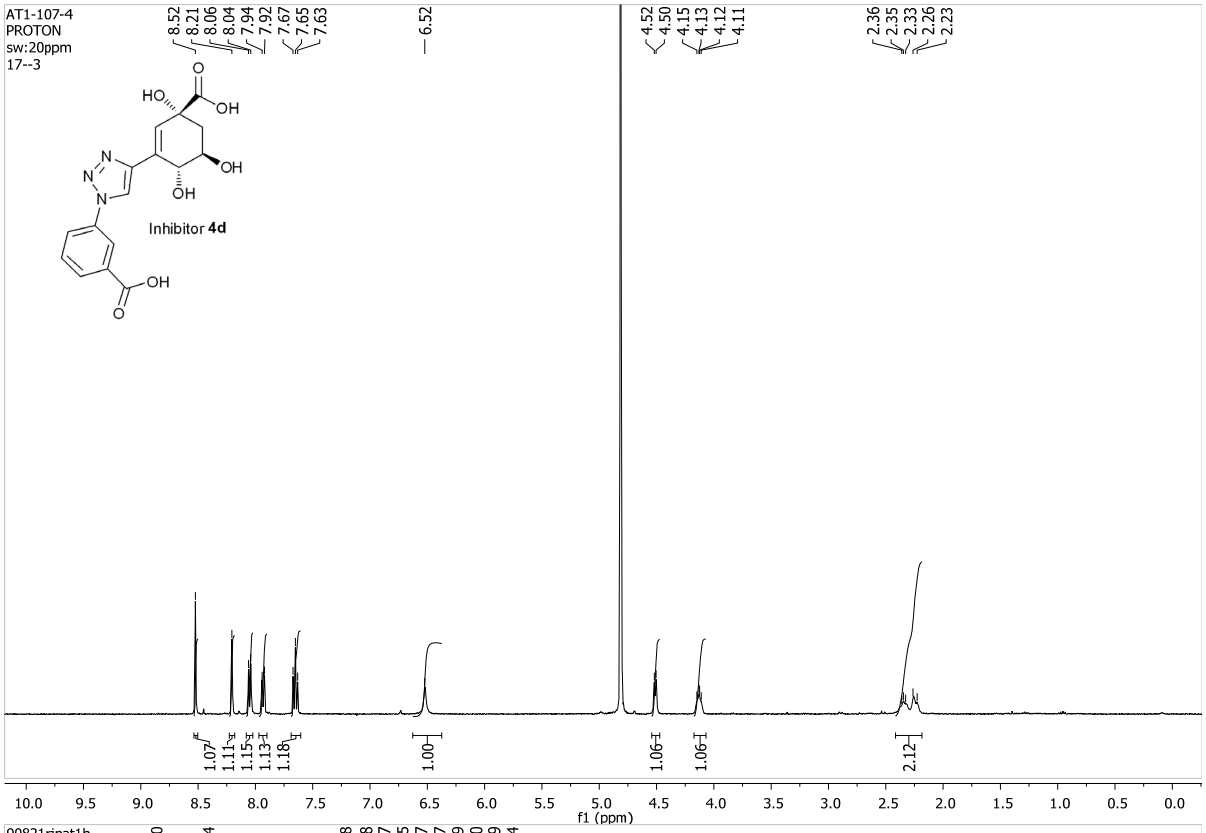

AT2-6
180.55
163.91
161.45
145.52
132.67
130.76
128.72
123.52
123.43
122.32
116.80
116.57
70.84
69.93
37.55
Inhibitor 4b
f1 (ppm)

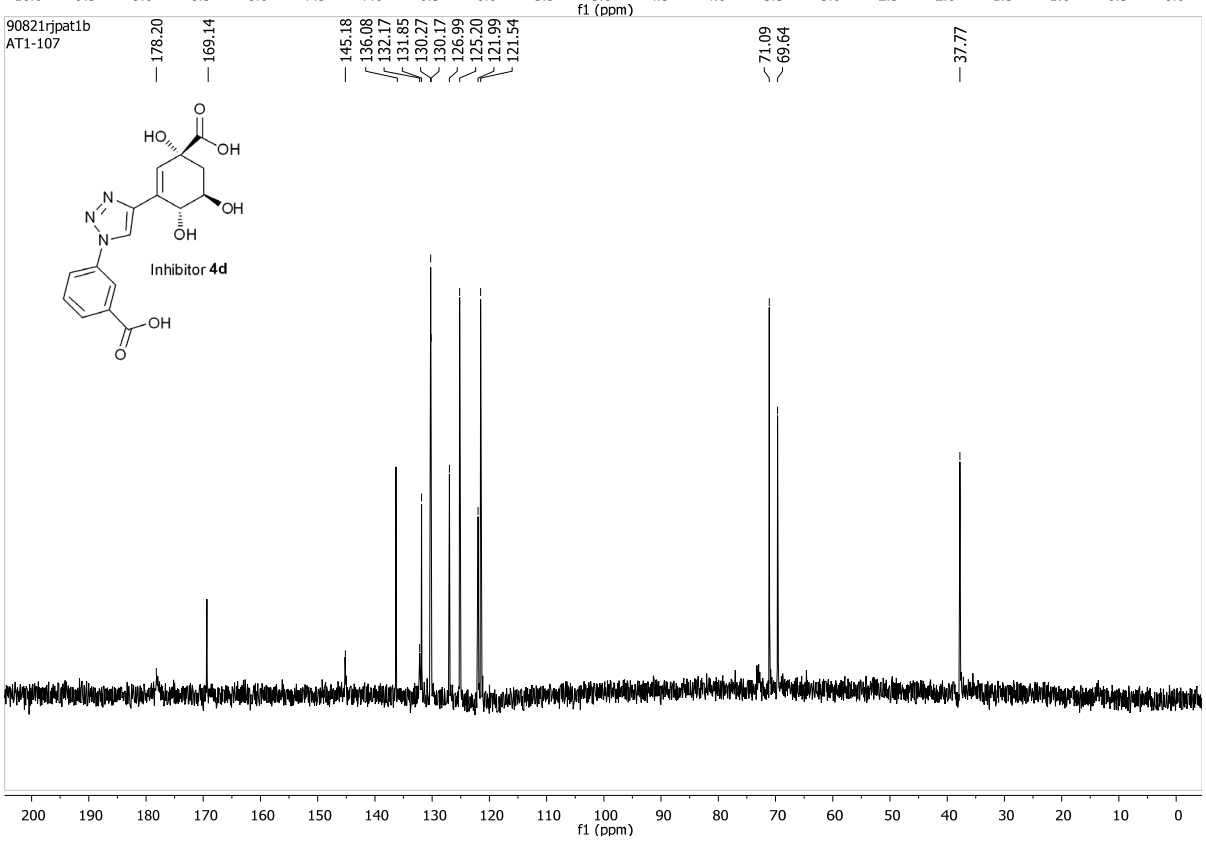

RT: 0.00 - 44.99 SM: 11G
Inhibitor 4b
NL:
3.68E5
Total Scan
PDA
at2-6final
uAU
Time (min)
15.84
2.23
2.06
3.62
3.98
8.43
11.82
12.98
13.37
14.71
17.60
20.83
22.84
24.43
26.09
27.13
28.78
31.14
32.03
33.79
35.79
37.41
38.63
40.20
41.19
43.83

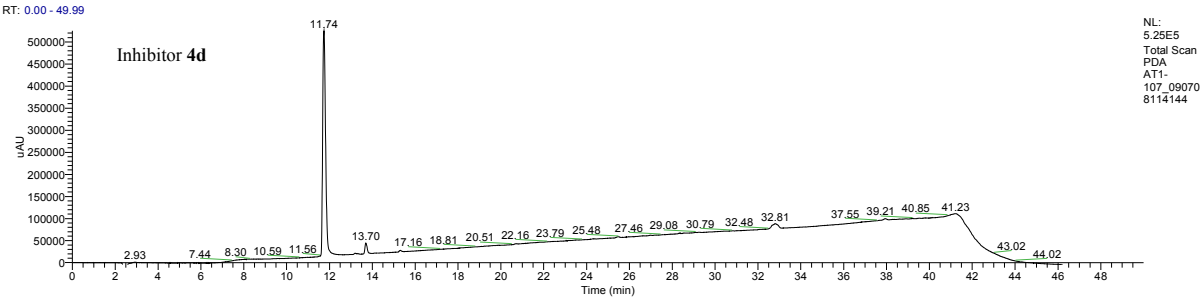

AT2-30-2
PROTON
sw:20ppm
17--3
Inhibitor 4c
f1 (ppm)

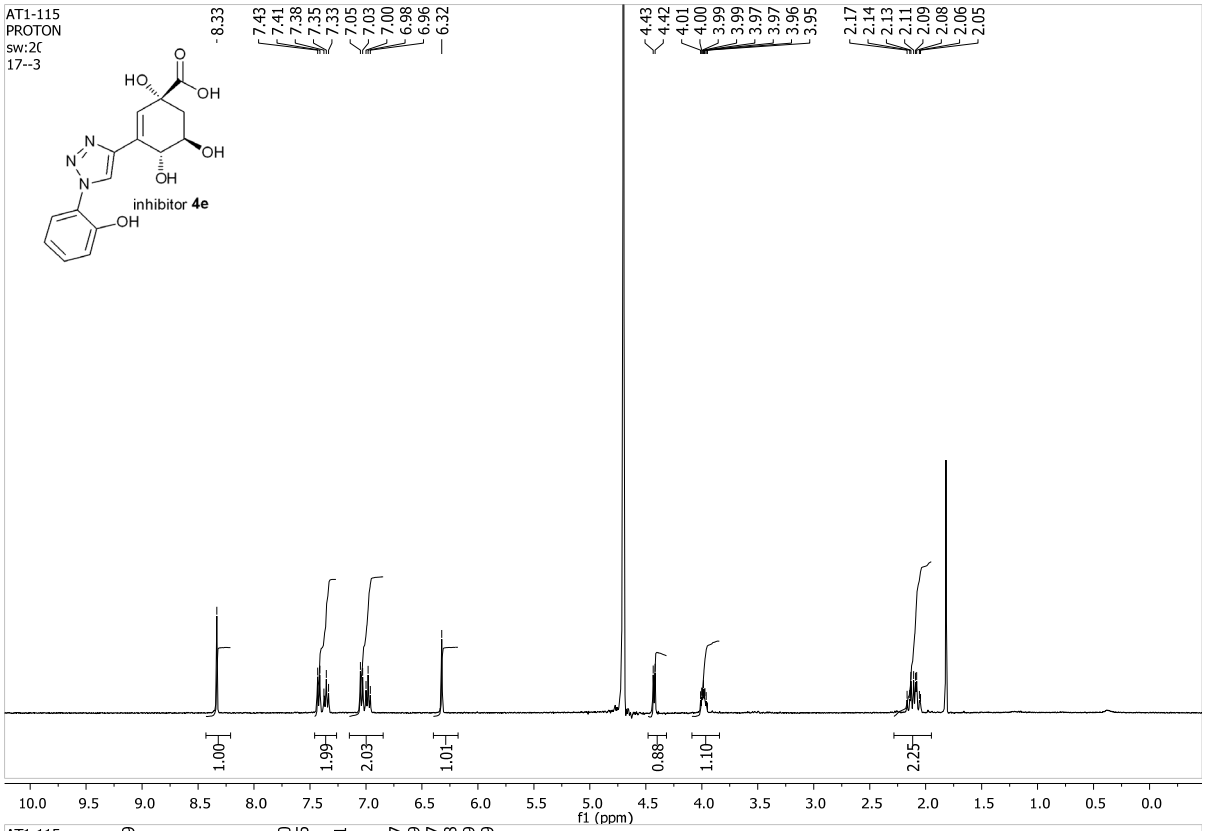

AT2-30-2
Inhibitor 4c
f1 (ppm)

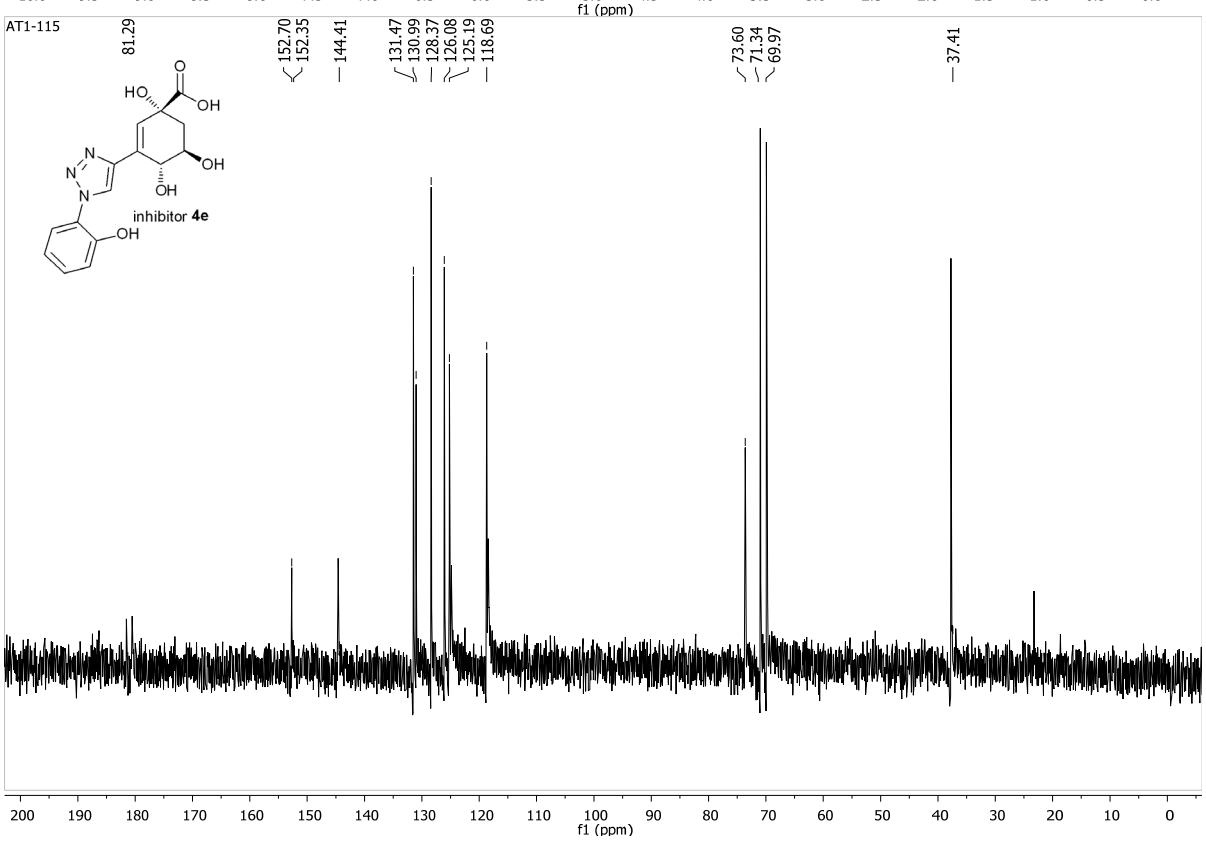

RT: 0.00 - 44.99 SM: 11G
Inhibitor 4c
NL:
3.89E5
Total Scan
PDA
AT2-30
uAU
Time (min)
15.95
1.78
2.33
3.56
4.63
8.48
11.08
12.71
14.26
15.63
16.49
17.32
21.07
22.38
24.23
25.97
27.63
29.09
29.27
32.38
33.73
35.61
37.33
38.51
40.04
41.15

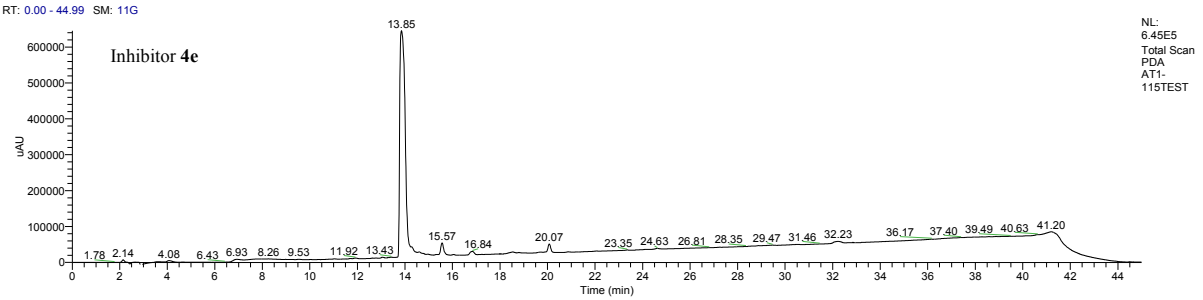

AT1-107-4
PROTON
sw:20ppm
17--3
8.52
8.21
8.06
8.04
7.94
7.92
7.67
7.65
7.63
6.52
4.52
4.50
4.15
4.13
4.12
4.11
2.36
2.35
2.33
2.26
2.23
Inhibitor 4d
1.07
1.11
1.15
1.13
1.18
1.00
1.06
1.06
2.12
f1 (ppm)

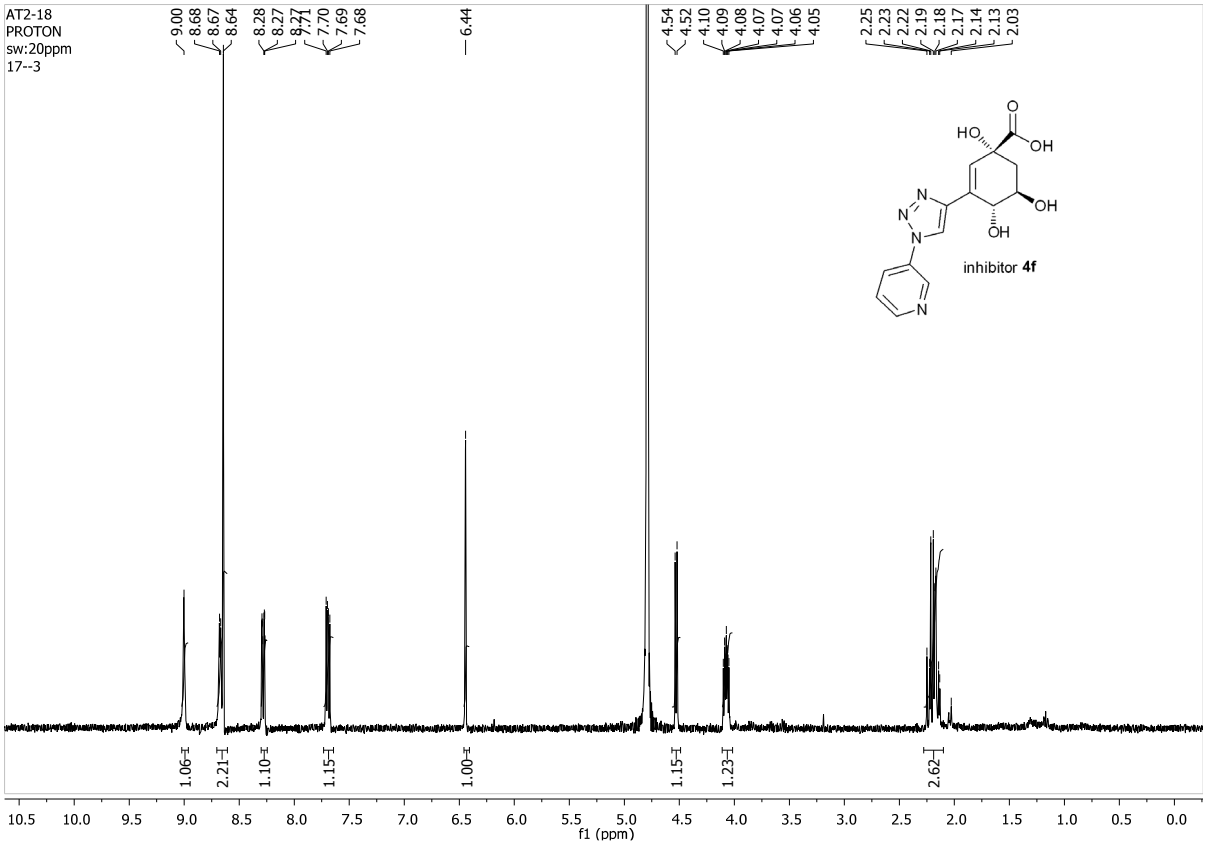

90821rjpat1b
AT1-107
178.20
169.14
145.18
136.08
132.17
131.85
130.27
130.17
126.99
125.20
121.99
121.54
71.09
69.64
37.77
Inhibitor 4d
f1 (ppm)

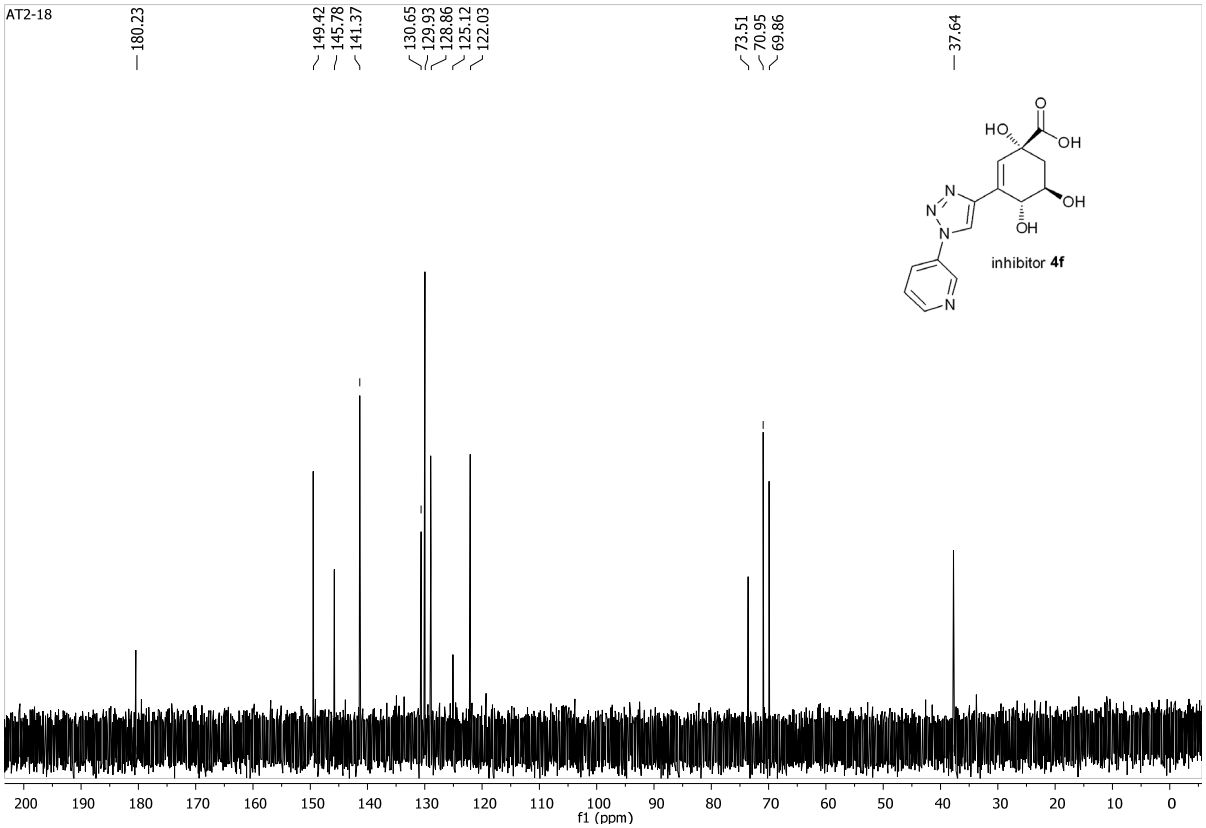

RT: 0.00 - 49.99
Inhibitor 4d
NL:
5.25E5
Total Scan
PDA
AT1-
107_09070
8114144
uAU
Time (min)
11.74
2.93
7.44
8.30
10.59
11.56
13.70
17.16
18.81
20.51
22.16
23.79
25.48
27.46
29.08
30.79
32.48
32.81
37.55
39.21
40.85
41.23
43.02
44.02

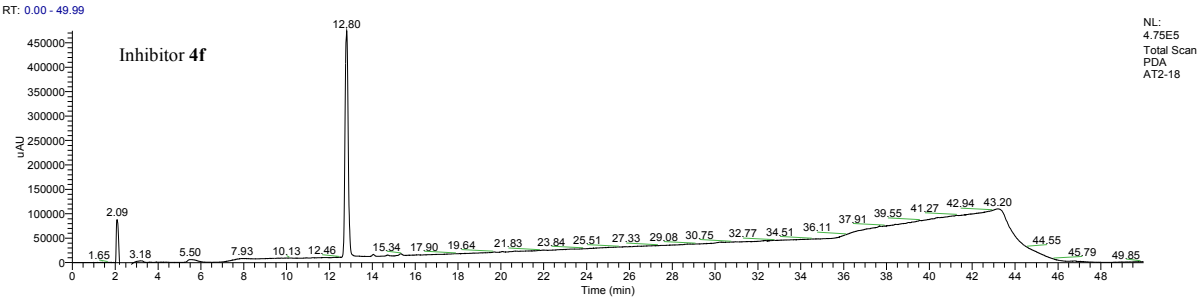

AT1-115
PROTON
sw:2(
17--3
inhibitor 4e
8.33
7.43
7.41
7.38
7.35
7.33
7.05
7.03
7.00
6.98
6.96
6.32
4.43
4.42
4.01
4.00
3.99
3.99
3.97
3.97
3.96
3.95
2.17
2.14
2.13
2.11
2.09
2.08
2.06
2.05
1.00
1.99
2.03
1.01
0.88
1.10
2.25
f1 (ppm)

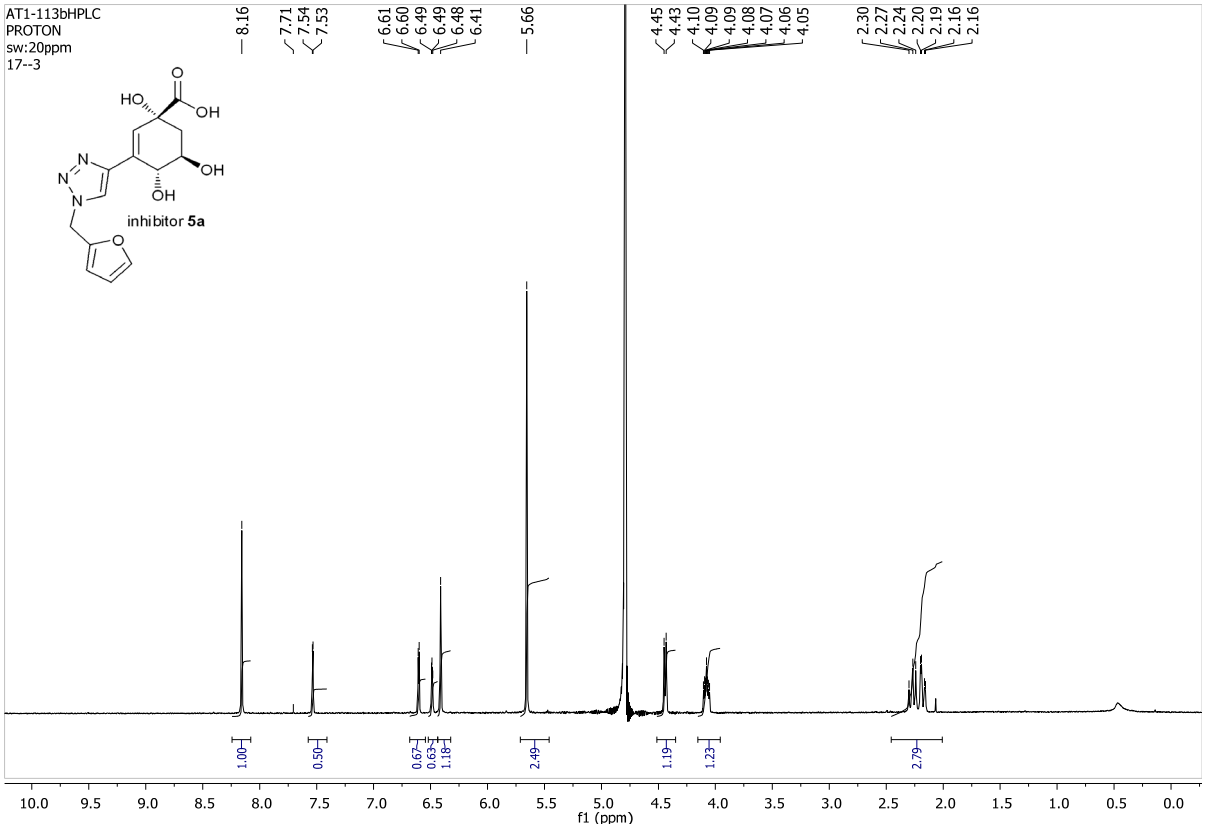

AT1-115
inhibitor 4e
81.29
152.70
152.35
144.41
131.47
130.99
128.37
126.08
125.19
118.69
73.60
71.34
69.97
37.41
f1 (ppm)

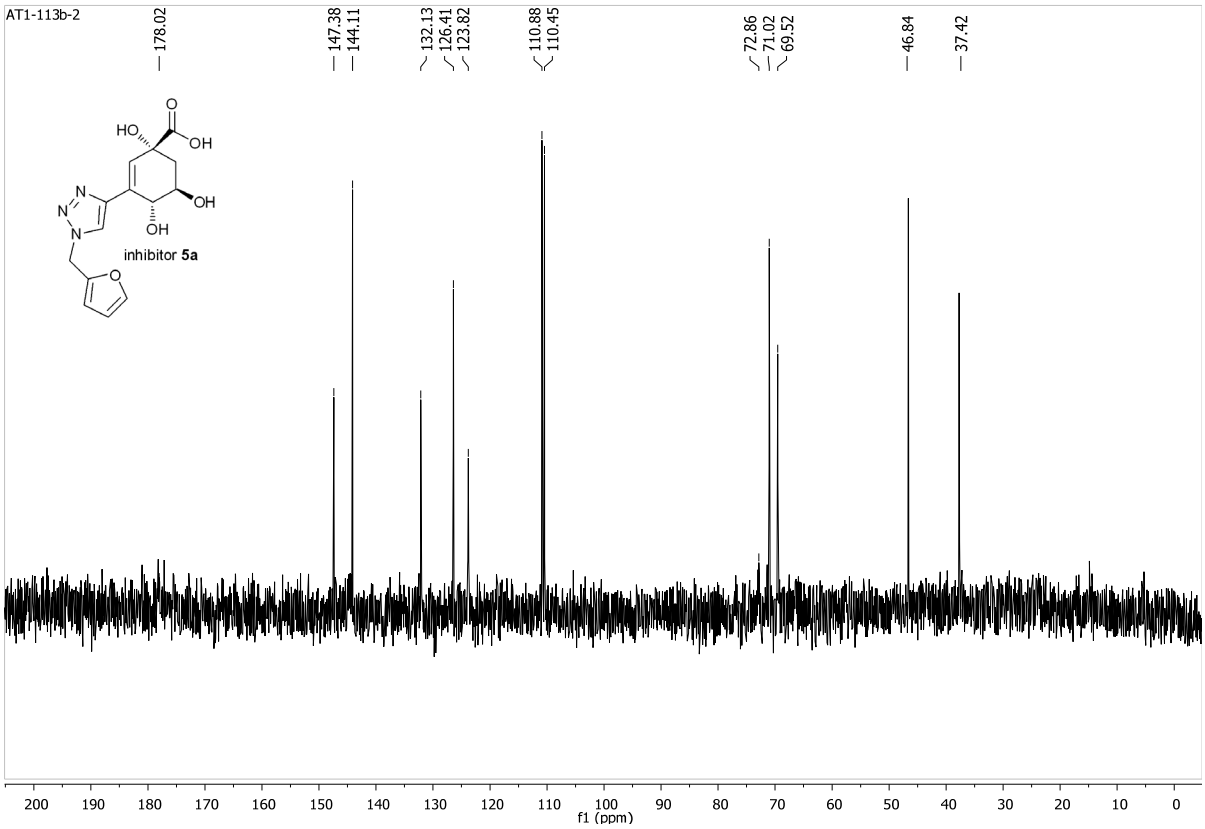

RT: 0.00 - 44.99 SM: 11G
Inhibitor 4e
NL:
6.45E5
Total Scan
PDA
AT1-
115TEST
uAU
Time (min)
13.85
1.78
2.14
4.08
6.43
6.93
8.26
9.53
11.92
13.43
15.57
16.84
20.07
23.35
24.63
26.81
28.35
29.47
31.46
32.23
36.17
37.40
39.49
40.63
41.20

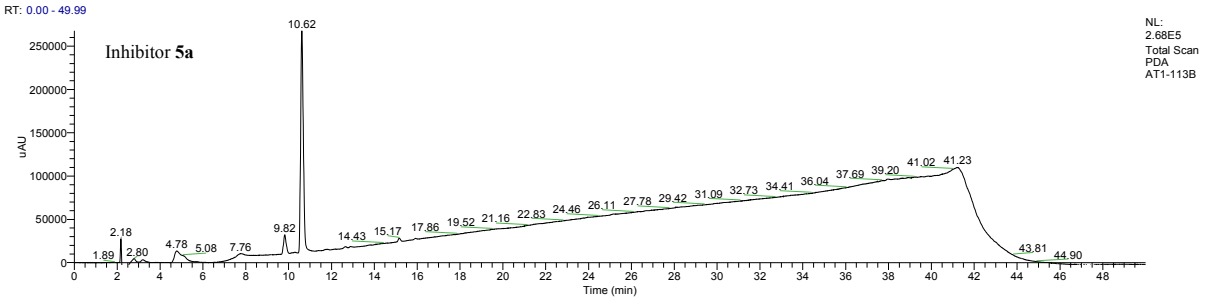

AT2-18
PROTON
sw:20ppm
17--3
9.00
8.68
8.67
8.64
8.28
8.27
7.71
7.70
7.69
7.68
6.44
4.54
4.52
4.10
4.09
4.08
4.07
4.07
4.06
4.05
2.25
2.23
2.22
2.19
2.18
2.17
2.14
2.13
2.03
1.06
2.21
1.10
1.15
1.00
1.15
1.23
2.62
HO
O
OH
OH
OH
N
N
N
N
inhibitor 4f
f1 (ppm)

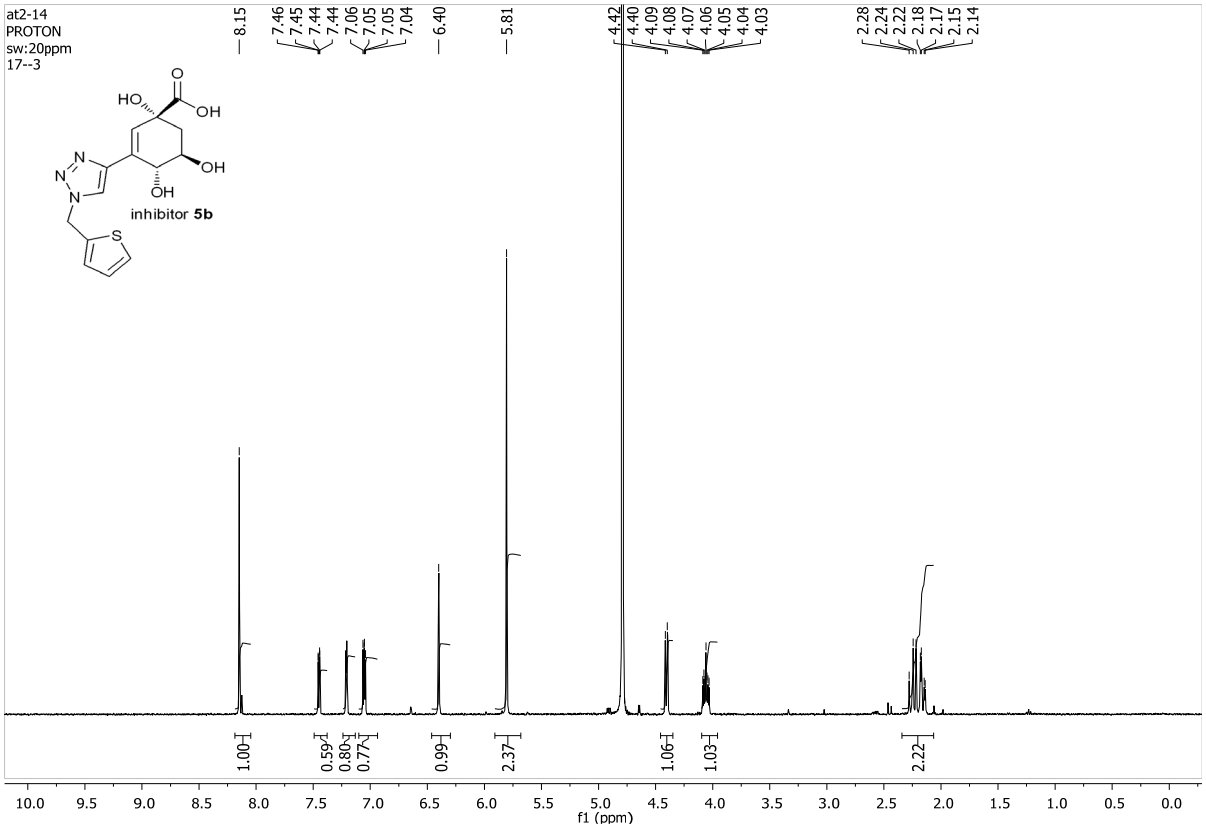

AT2-18
180.23
149.42
145.78
141.37
130.65
129.93
128.86
125.12
122.03
73.51
70.95
69.86
37.64
HO
O
OH
OH
OH
N
N
N
N
inhibitor 4f
f1 (ppm)

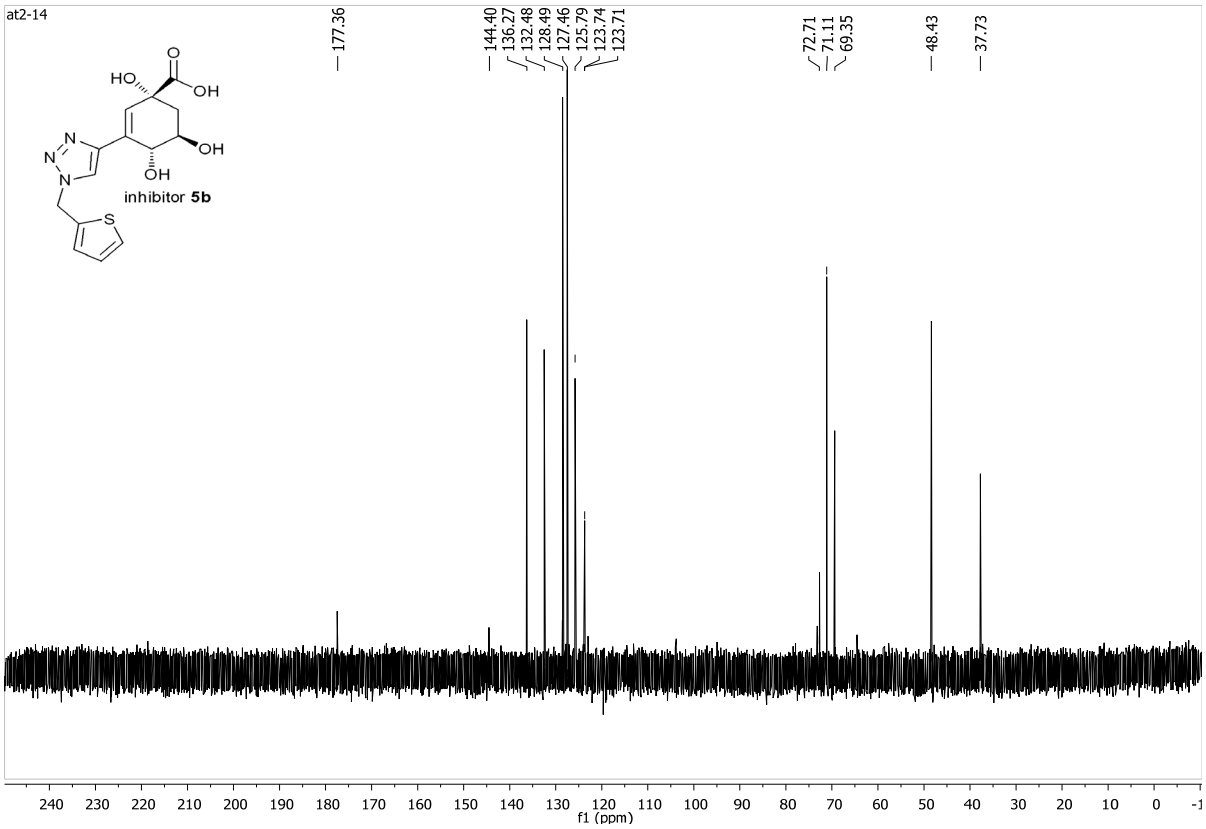

RT: 0.00 - 49.99
Inhibitor 4f
NL:
4.75E5
Total Scan
PDA
AT2-18
uAU
Time (min)
12.80
2.09
1.65
3.18
5.50
7.93
10.13
12.46
15.34
17.90
19.64
21.83
23.84
25.51
27.33
29.08
30.75
32.77
34.51
36.11
37.91
39.55
41.27
42.94
43.20
44.55
45.79
49.85

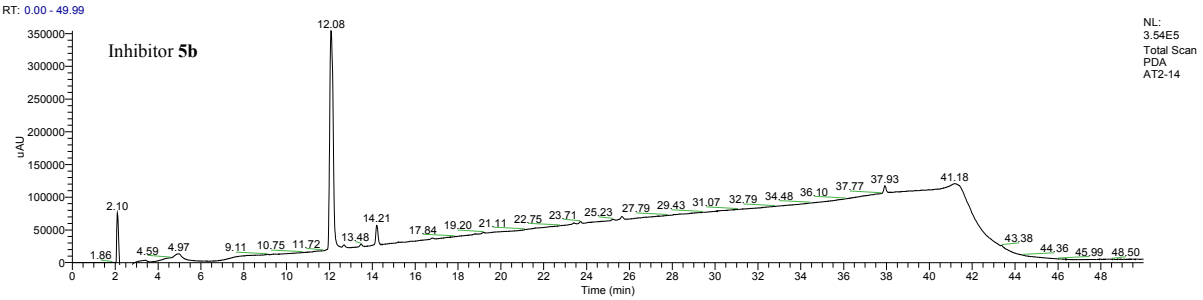

AT1-113bHPLC
PROTON
sw:20ppm
17--3
8.16
7.71
7.54
7.53
6.61
6.60
6.49
6.49
6.48
6.41
5.66
4.45
4.43
4.10
4.09
4.09
4.08
4.07
4.06
4.05
2.30
2.27
2.24
2.20
2.19
2.16
2.16
inhibitor 5a
1.00
0.50
0.67
0.63
1.18
2.49
1.19
1.23
2.79
f1 (ppm)


AT1-113b-2
178.02
147.38
144.11
132.13
126.41
123.82
110.88
110.45
72.86
71.02
69.52
46.84
37.42
inhibitor 5a
f1 (ppm)


RT: 0.00 - 49.99
Inhibitor 5a
NL:
2.68E5
Total Scan
PDA
AT1-113B
uAU
Time (min)
1.89
2.18
2.80
4.78
5.08
7.76
9.82
10.62
14.43
15.17
17.86
19.52
21.16
22.83
24.46
26.11
27.78
29.42
31.09
32.73
34.41
36.04
37.69
39.20
41.02
41.23
43.81
44.90


at2-14
PROTON
sw:20ppm
17--3
inhibitor 5b
f1 (ppm)


at2-14
inhibitor 5b
f1 (ppm)


RT: 0.00 - 49.99
Inhibitor 5b
NL:
3.54E5
Total Scan
PDA
AT2-14
uAU
Time (min)
1.86
2.10
4.59
4.97
9.11
10.75
11.72
12.08
13.48
14.21
17.84
19.20
21.11
22.75
23.71
25.23
27.79
29.43
31.07
32.79
34.48
36.10
37.77
37.93
41.18
43.38
44.36
45.99
48.50

References

1. A. W. Roszak, D. A. Robinson, T. Krell, I. S. Hunter, M. Fredrickson, C. Abell, J. R. Coggins and A. J. Lapthorn, *Structure*, 2002, **10**, 493-503.

2. V. F. V. Prazeres, L. Tizón, J. M. Otero, P. Guardado-Calvo, A. L. Llamas-Saiz, M. J. van Raaij, L. Castedo, H. Lamb, A. R. Hawkins and C. González-Bello, *J. Med. Chem.*, 2010, **53**, 191-200.

3. R. A. Friesner, R. B. Murphy, M. P. Repasky, L. L. Frye, J. R. Greenwood, T. A. Halgren, P. C. Sanschagrin and D. T. Mainz, *J. Med. Chem.*, 2006, **49**, 6177-6196.

4. F. W. Studier, *Protein Expr. Purif.*, 2005, **41**, 207-234.

5. L. D. B. Evans, A. W. Roszak, L. J. Noble, D. A. Robinson, P. A. Chalk, J. L. Matthews, J. R. Coggins, N. C. Price and A. J. Lapthorn, *FEBS Lett.*, 2002, **530**, 24-30.

6. V. F. V. Prazeres, C. Sánchez-Sixto, L. Castedo, H. Lamb, A. R. Hawkins, A. Riboldi-Tunnicliffe, J. R. Coggins, A. J. Lapthorn, C. González-Bello, *ChemMedChem*, 2007, **2**, 194-207.

7. K. Barral, A. D. Moorhouse, J. E. Moses, *Org. Lett.* 2007, **9**, 1809-1811.

8. K. N. Crabtree, K. J. Hostetler, T. E. Munsch, P. Neuhaus, P. M. Lahti, W. Sander, J. S. Poole, *J. Org. Chem.* 2008, **73**, 3441-3451.

9. S. A. Rogers, C. Melander , *Angew. Chem. Int. Ed.* 2008, **47**, 5229-5231.

10. E. D. Goddard-Borger, R. V. Stick, *Org. Lett.* 2007, **9**, 3797-3800.