A Unique Molecular Interaction with Histone Deacetylase 6 Catalytic Tunnel: Crystallographic and Biological Characterization of a Model Chemotype

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Supplementary Information

Fig S1A: Docking pose of **1** in danio rerio HDAC6 catalytic domain 2, CD2 (*dr*HDAC6, **PDB 5WGI**) binding pocket. Full description of in silico experiments are detailed out below in supplementary methods under computational modelling/docking studies.

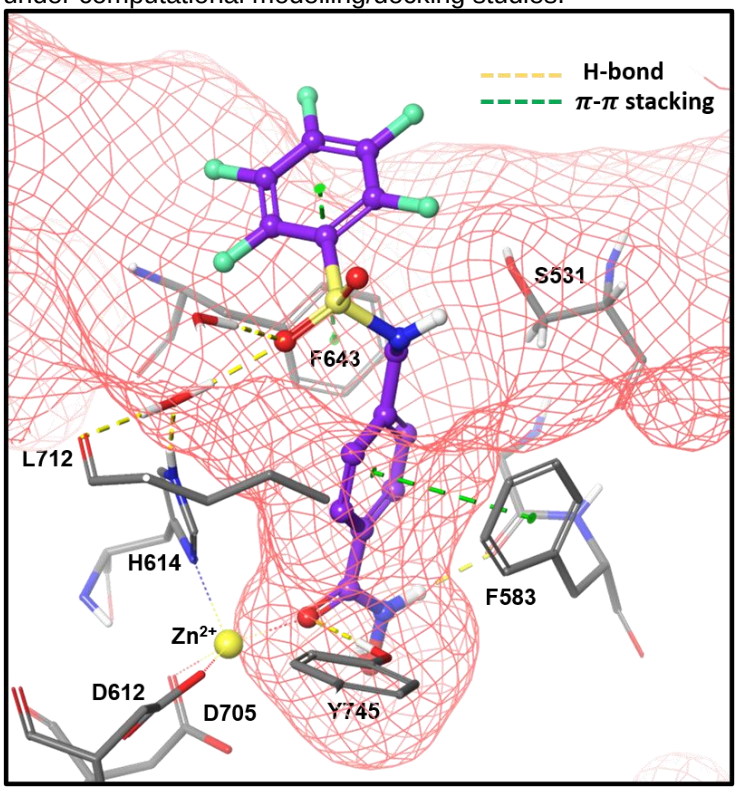


Fig. S1B: Aerial view of hHDAC6 (**PDB 5EDU**) CD2 showing the L1 & L2 pockets and their relevant associated residues.

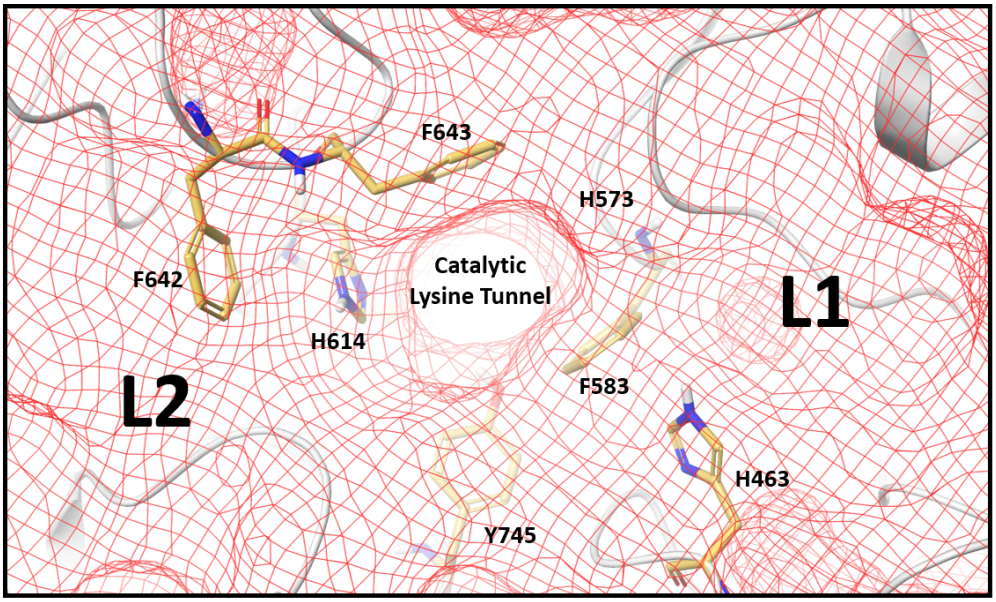


Fig. S1C: Approximate dimensions of the active site tunnel for HDAC3 (**PDB: 4A69**), HDAC6 (**PDB: 5WGI**), and HDAC8 (**PDB: 1T64**). Approximate dimensions were determined using the built-in distance calculator on the Schrödinger Maestro 12.2.012 software

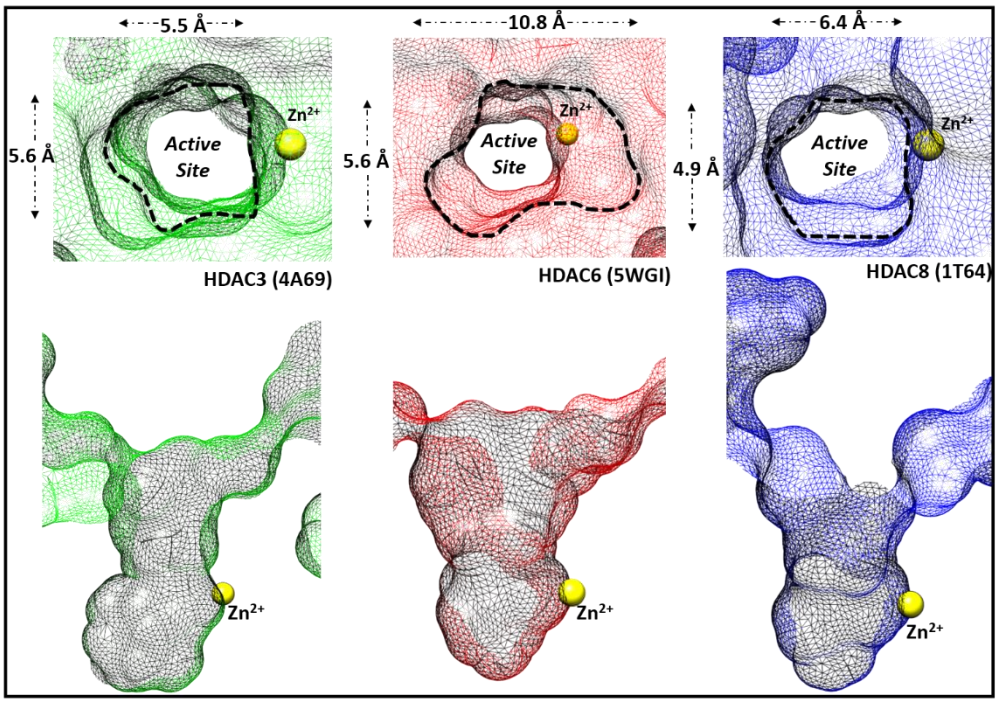


Fig. S2: Docking pose of **2** at the *dr*HDAC6 (**PDB 5WGI**) CD2. The aromatic substitution seems to capture more interactions at the catalytic surface, mainly pi-pi aromatic interactions with F643

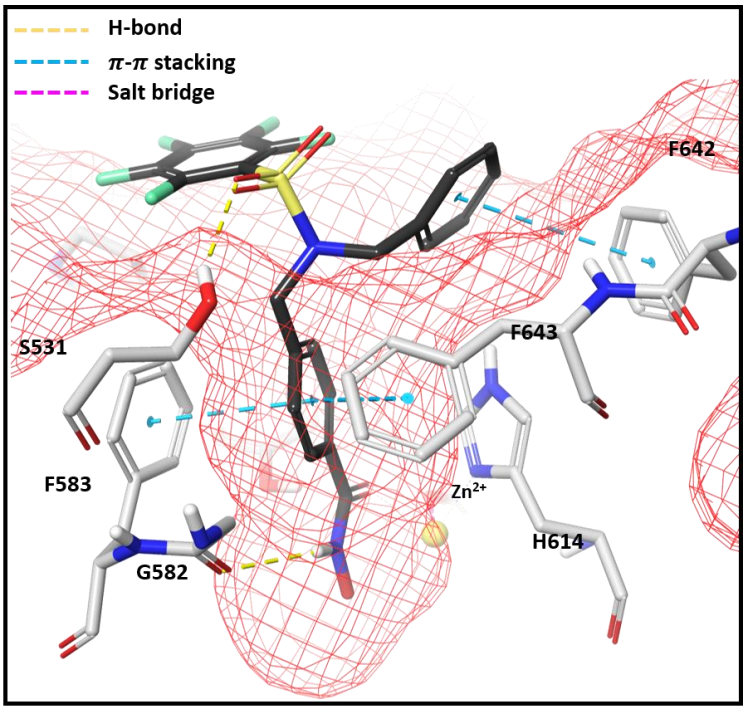


Fig. S3: Fluorescence Polarization (FP) assay shows superior potency of TO-317 in the library of compounds evaluated against HDAC6 and corroborates initial EMSA IC_{50} data presented in main article.

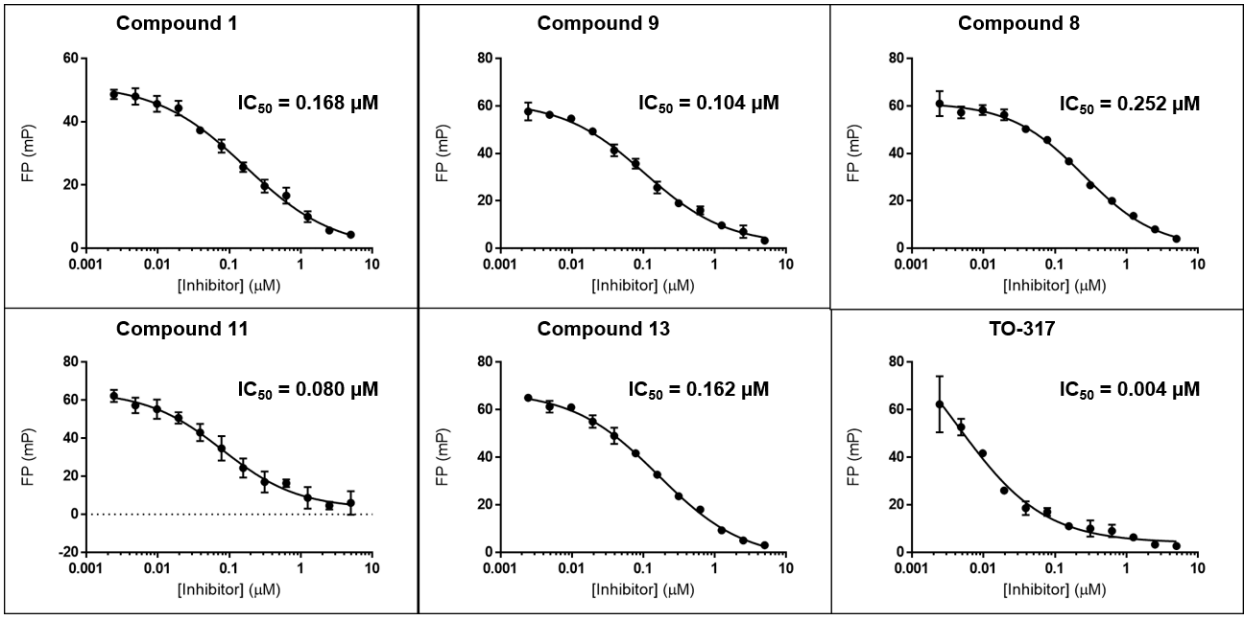


Fig. S4-A: An overlay of the docking pose of Citarinostat (with *dr*HDAC6 CD2, **PDB 5WGI**) with a crystal image of ricolinostat with *dr*HDAC6 (**PDB 5WGL**)

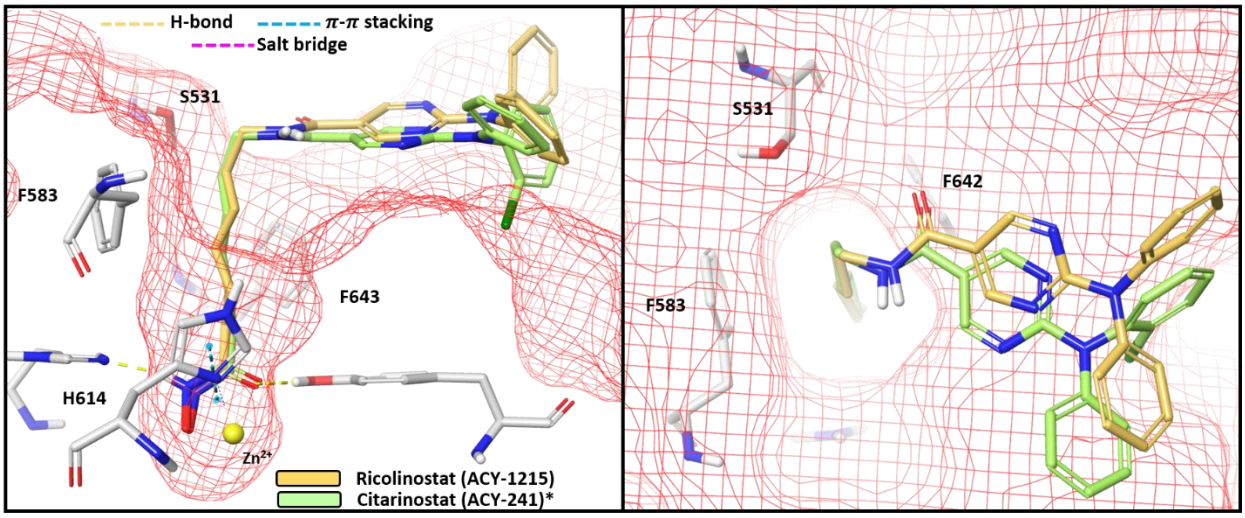


Fig. S4-B: Docking poses of TO-317 with *human* HDAC8, *h*HDAC8 (**PDB 1T64**). Deck A shows binding pocket view with minimal residue engagements at the catalytic tunnel, deck B shows aerial view revealing a solvent exposed pyridyl motif, unlike within the drHDAC6 pocket where notable hydrogen bonding interactions with the Zn^{2+}-ligand, H614, were observed

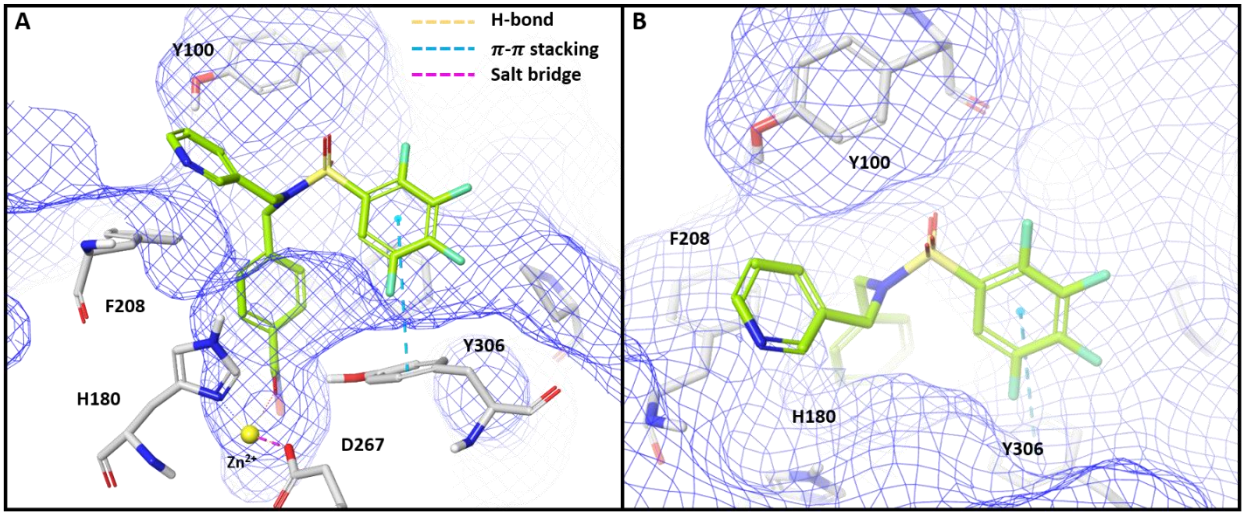


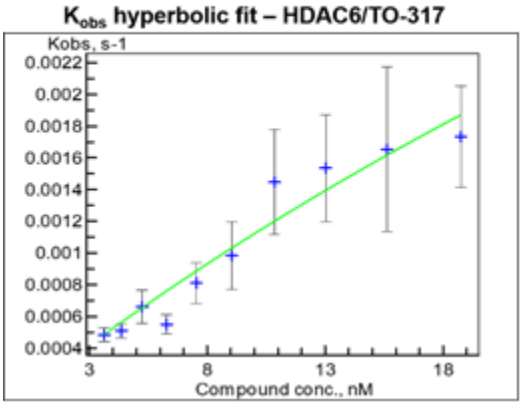
Table S1: Comparison of **i**n silico ΔG_B calculation of the binding of some selected inhibitors with HDAC6 & HDAC8

In silico **ΔG_B (kcal/mol)**	
Compound	**HDAC6**
Citarinostat	-7.939
A	-8.843
1	-8.464
TO-317	-9.065

In silico **ΔG_B (kcal/mol)**		
Compound	**HDAC6**	**HDAC8**
TO-317	-9.065	-7.565

Fig. S5-A: Kinetic analysis of TO-317/HDAC6 complex formation. A two-step reversible model was employed based on a jump-dilution kinetic assay. Full details on experimental procedures are provided below in supplementary methods under in vitro HDAC IC_{50} determination.

Two-step reversible model ($k_{-2} > 0$) $E + I \rightleftharpoons E:I \rightleftharpoons E:I^*$

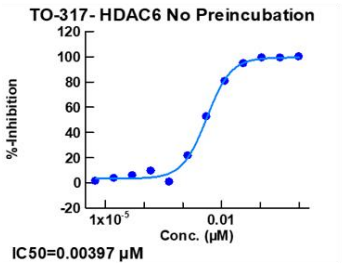


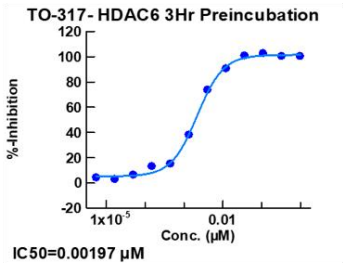
$K_{obs} = K_{-2} + K2^*[I]/([I] + Ki^*)$

Parameter	Value
K_2 (s^{-1})	0.01
Ki*(1+[S]/Km) (nM)	87.03
K_{-2} (S^{-1})	8.14E-05
Ki-initial (nM)	**87.0**
Ki-overall (nM)	**0.70**
Residence Time (min)	**142**
Chi^2	1.56E-07
F-test	0.83

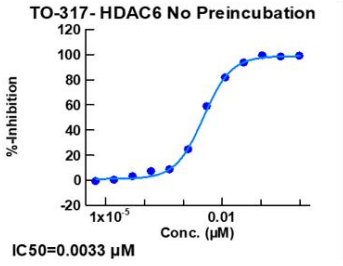
Fig. S5-B: Incubation IC_{50} Studies for TO-317's Inhibition of full-length *human* HDAC6, *h*HDAC6.

Run	Compound	HDAC6 No Pre-incubation (IC_{50}, µM)	HDAC6 3h Pre-incubation (IC_{50}, µM)
1	TO-317	0.00397	0.00197





Run	Compound	HDAC6 No Pre-incubation (IC_{50}, µM)	HDAC6 3h Pre-incubation (IC_{50}, µM)
2	TO-317	0.0033	0.00178



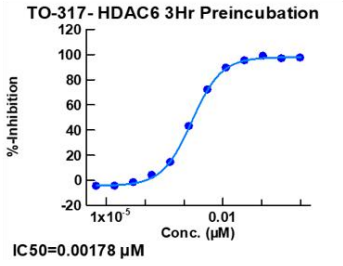


Fig. S5-C: Mass Spectrometry Data for TO-317 with HDAC6

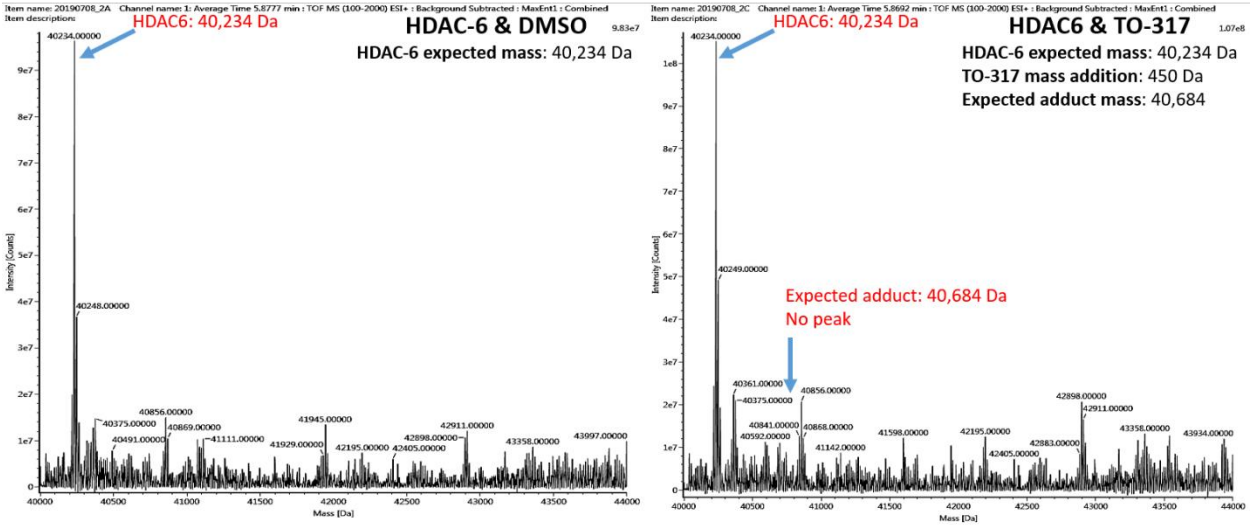


Fig. S6: Nuclear stain of HeLa cells treated with TO-317 and Citarinostat shown in blue. Overlay of both Ac-α-tubulin stain (red)/nuclear stain and Ac-α-tubulin stain/Ac-H3K27 histones are shown.

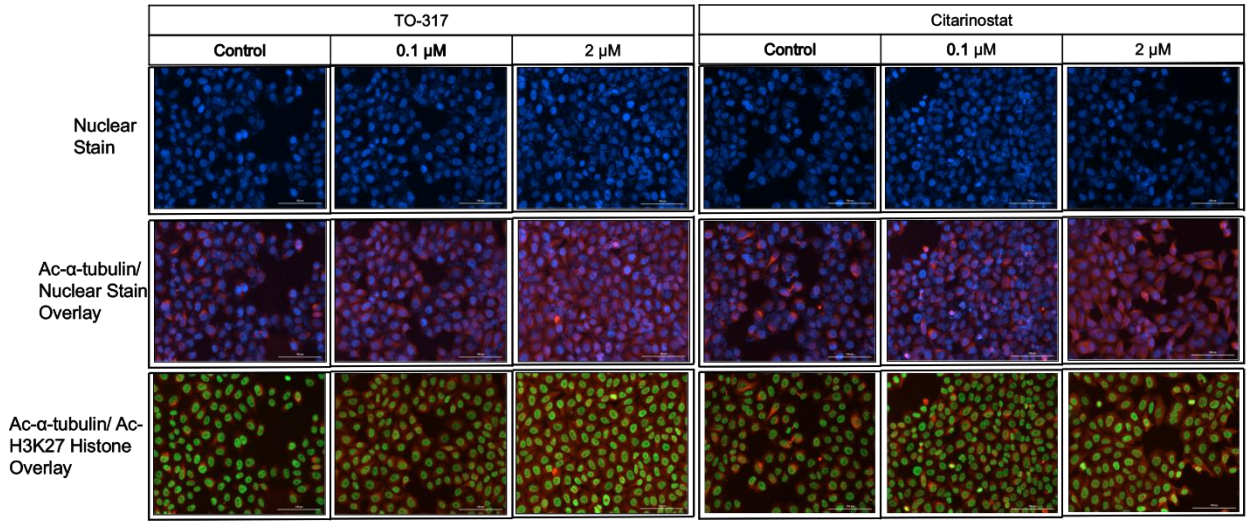


Fig. S7: HeLa cells with TO-317 and Citarinostat at two doses for 6 h. Red – acetylated α-tubulin; Green – acetylated histones. Fluorescence imaging shows TO-317 treated HeLa cells leads to drastic accumulation of acetylated α-tubulin at 2 μM without significant accumulation of acetylated histones. Images from Citarinostat stain shows modest build-up of α-tubulin at 2 μM, which is of less intensity compared to TO-317 at the same concentration.

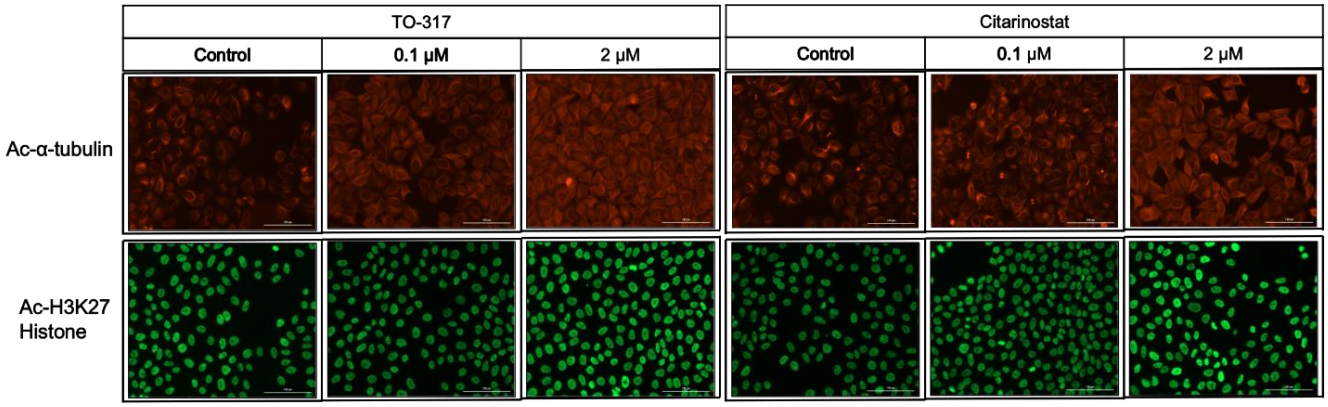


Fig. S8-A: Population sorting of MV4-11 cells at different concentrations of TO-317. A dose-dependent increase in apoptotic cells were observed when TO-317 was incubated with MV4-11 cells for 6 h revealing a mechanistic approach to the anti-proliferative activity of TO-317 in several cancer cells.

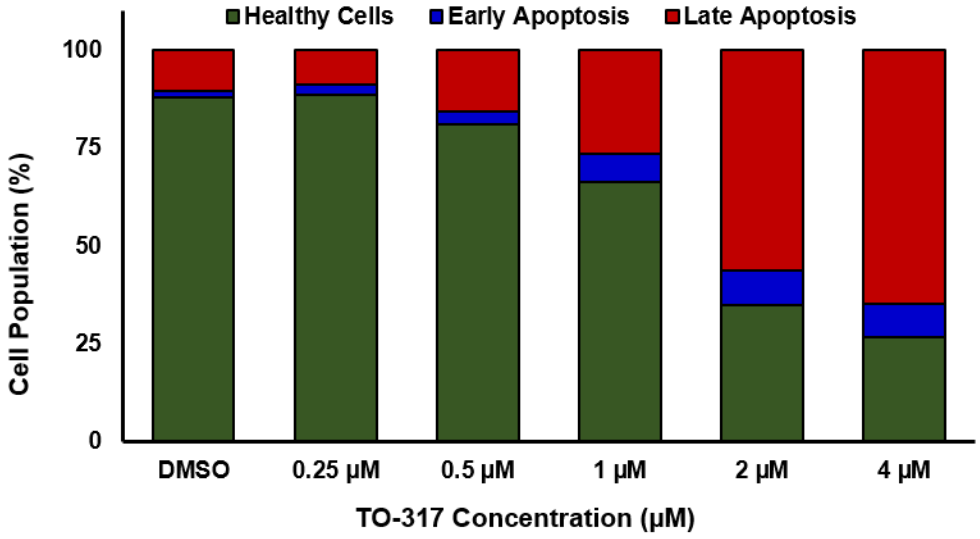


Fig. S8-B: Population sorting of MV4-11 cells at different concentrations of Citarinostat. Although a similar dose-dependent increase in apoptotic cells were observed when Citarinostat was incubated with MV4-11 cells for 6 hrs, it shows weaker potency when compared to TO-317.

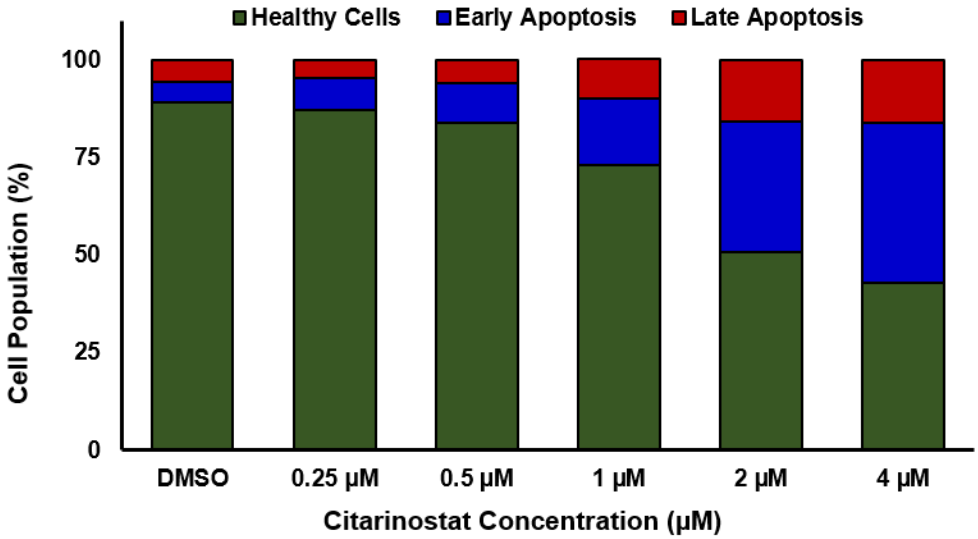
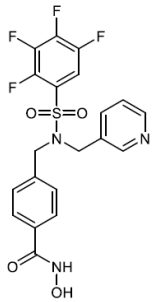


Fig. S9: Kinetic solubility plots of TO-317 in phosphate buffer solution (PBS) at pH7.4, 5% DMSO. HPLC quantification was used after filtration at each concentration studied.

- **TO-317**
  - **Saturation concentration (PBS pH 7.4, 5% DMSO):**
    - **161.50 ± 33.20 µM (n = 3)**
    - **142.35 ± 2.40 µM (n = 2)**



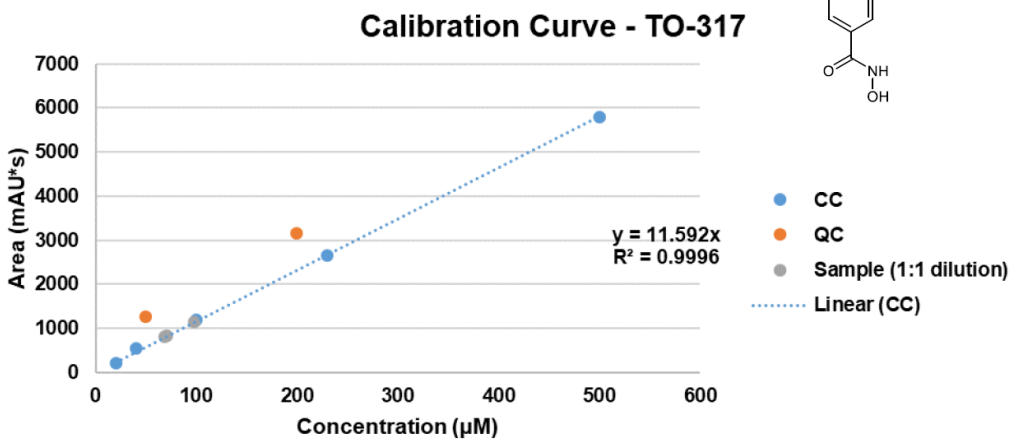


Table S2: TO-317 permeability determination by Lipid-PAMPA method

Compound	Replicate 1	Replicate 2	Replicate 3	Average Score	% Recovery
TO-317	6.53	6.96	6.61	6.71	111.5%

Scores are reported as -logP_e scores. The compounds can be categorized into high and low permeability. Compounds which have a -Log P_e < 6 are classified as high permeability and compounds with a -Log P_e > 6 are classified as low permeability.

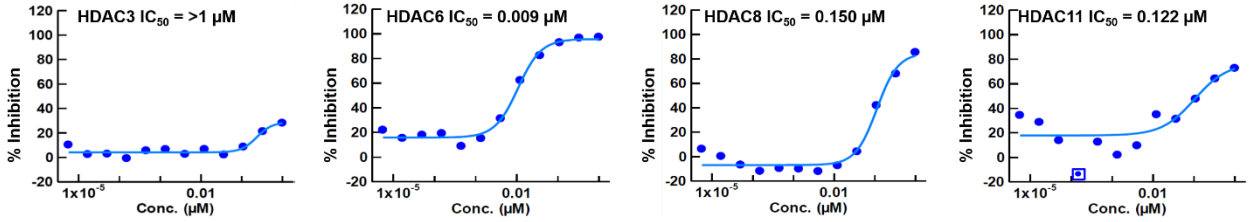
Table S3: Cell viability assays in different cell lines

Cell Lines	TO-317 IC_{50} (µM)	Citarinostat IC_{50} (µM)
RPMI8226	0.7 *(±0.06)*	6.5 *(±0.22)*
MM.1S	0.7 *(±0.06)*	1.6 *(±0.16)*
U87G	2.0 *(±0.15)*	9.1 *(NA)*
MOLM-13	2.0 *(±0.21)*	13.5 *(±1.66)*
MV4-11	0.6 *(±0.07)*	1.2 *(±0.47)*
Jurkat	1.9 *(±0.11)*	1.2 *(±0.16)*
AML3	3.3 *(±0.41)*	7.7 *(±1.59)*
AR230	1.5 *(NA)*	4.3 *(±2.02)*
BV-173	1.7 *(±0.19)*	0.9 *(±0.13)*
Ramos	0.3 *(±0.06)*	ND
HeLa	3.5 *(±1.43)*	7.0 *(±2.06)*
MRC-9	>50	30.4 *(±7.95)*

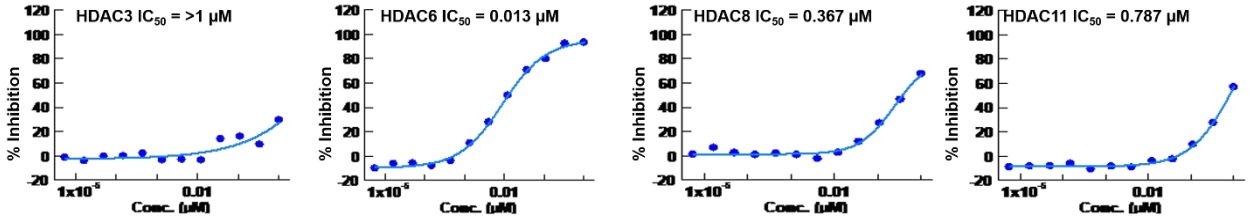
95% confidence interval are reported in brackets after each IC_{50}
NA – Not applicable, 95% confidence interval too wide
ND – Not determined

Fig. S10 IC_{50} Curves for HDAC inhibition determined by EMSA-based activity assays

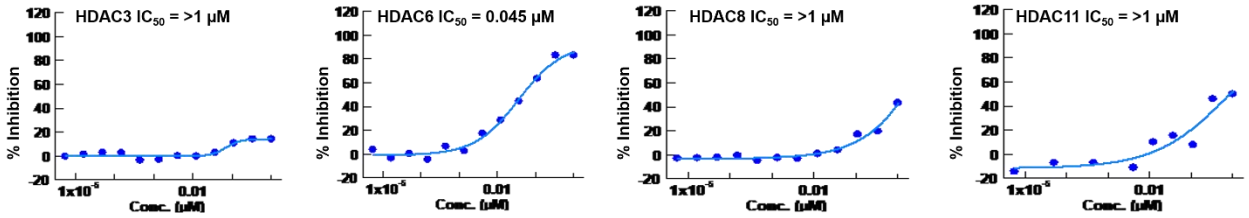
Compound **2** IC_{50} Curves



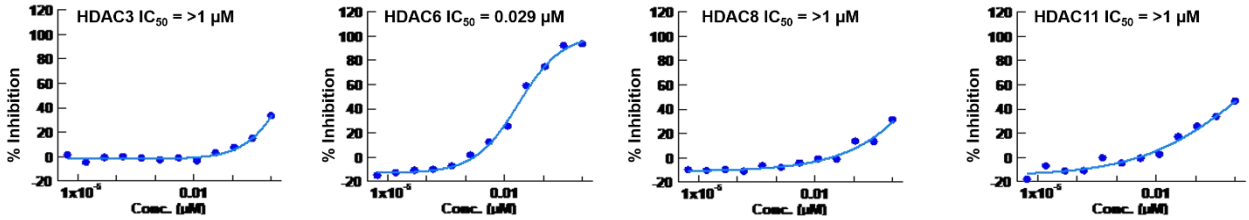
Compound **3** IC_{50} Curves



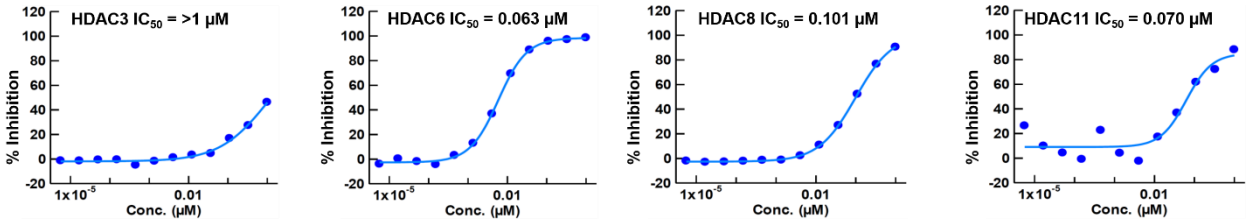
Compound **4** IC_{50} Curves



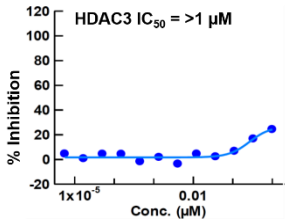
Compound **5** IC_{50} Curves

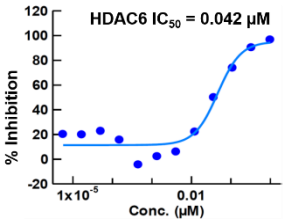


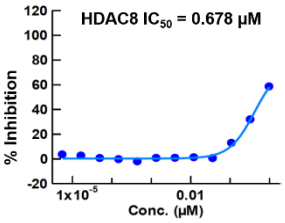
Compound **6** IC_{50} Curves

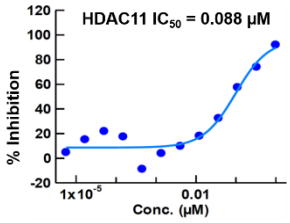


Compound **7** IC_{50} Curves









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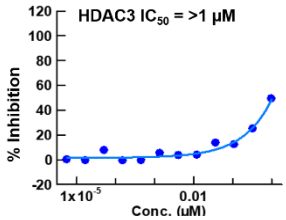
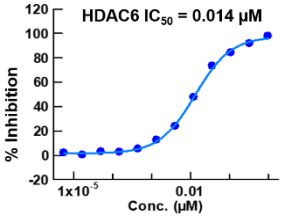
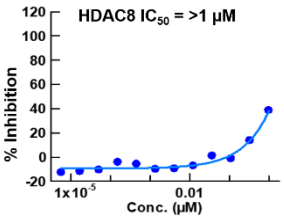
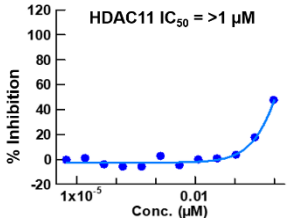
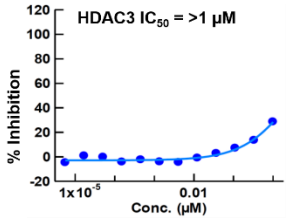


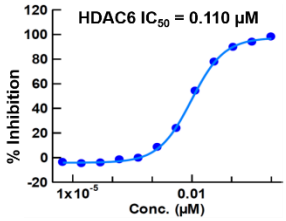


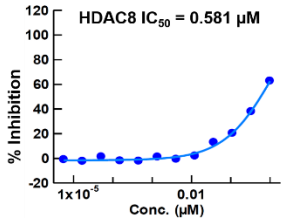


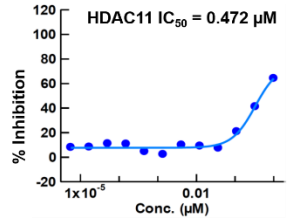


Compound **9** IC_{50} Curves

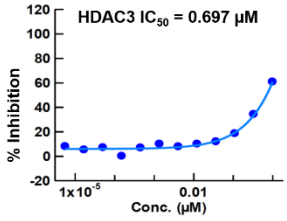


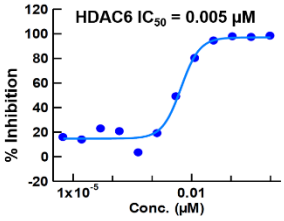


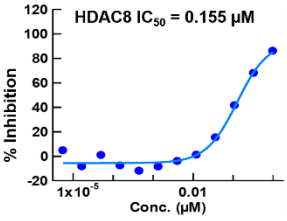


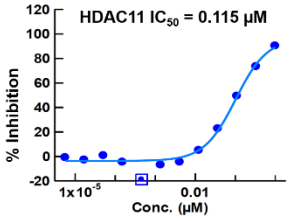


Compound **10** IC_{50} Curves

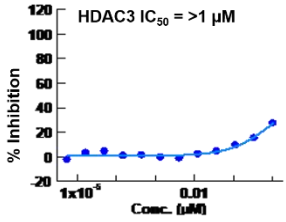


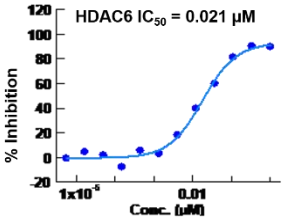


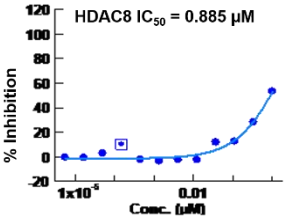


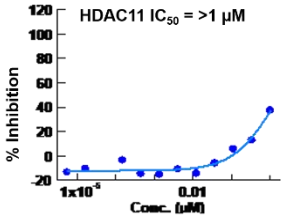


Compound **11** IC_{50} Curves

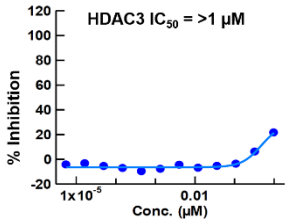


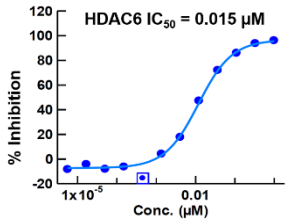


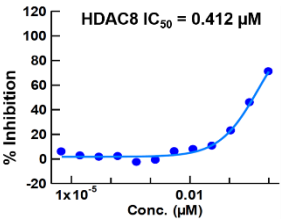


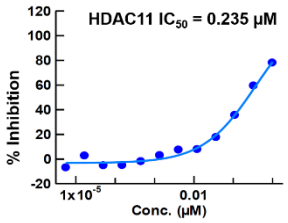


Compound **12** IC_{50} Curves

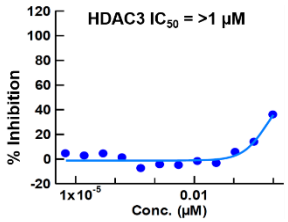


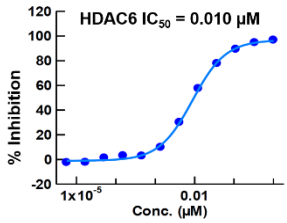


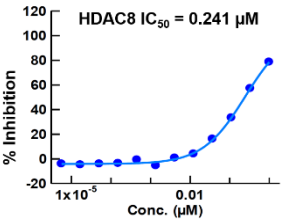


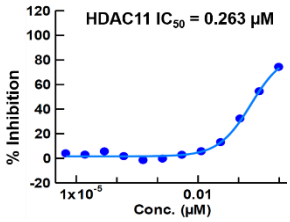


Compound **13** IC_{50} Curves

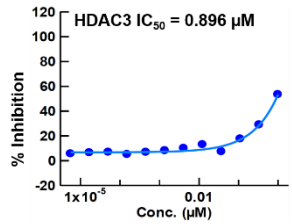


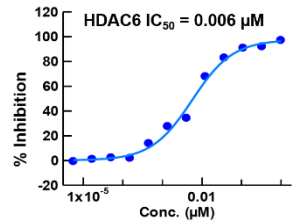


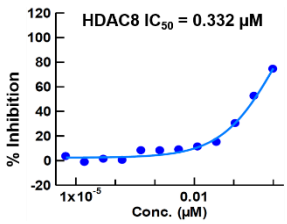


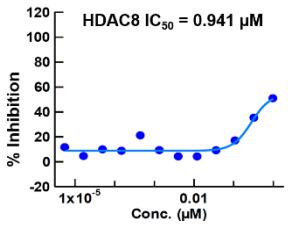


Compound **14** IC_{50} Curves

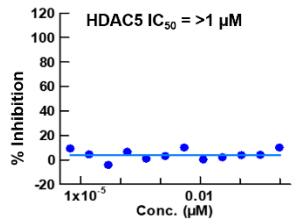


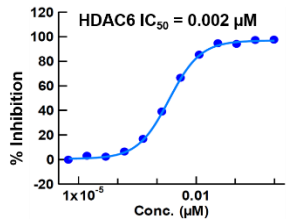


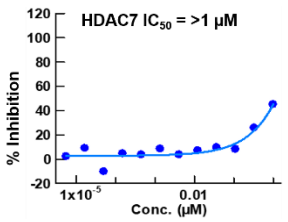


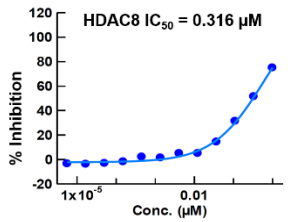


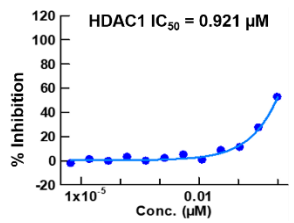
Compound **15 (TO-317)** IC_{50} Curves

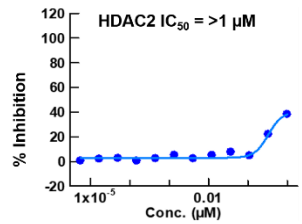


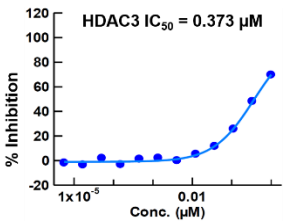


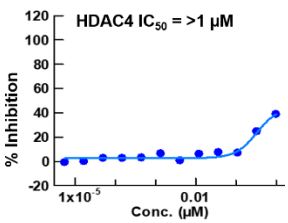


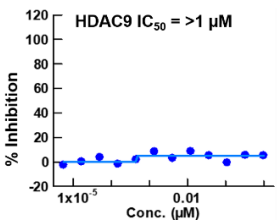


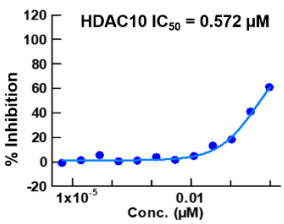


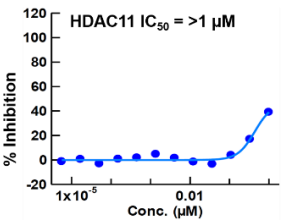












Supplementary Methods

Computational Modelling/Docking Studies

All described *in silico* experiments were performed on the Schrödinger Maestro 12.2.012 software using Glide within the Small-Molecule Drug Discovery Suite 2019-1 (Schrödinger, LLC, New York, NY, 2019).[1–3] These studies required the following programs within the above suite: Epik, Optimized Potentials for Liquid Simulations 3e (OPLS3e) force-field, Glide, LigPrep and the Protein Preparation Wizard. Other software used include ChemDraw Professional 17.1. All ligand poses and protein structure images were generated with Maestro 12.2.012.

Ligand Structure Preparation

The chemical structure of the HDACi were manually drawn into Maestro 12.2.012 using the 2D sketcher functionality, before undergoing a ligand preparation workflow (LigPrep, Schrödinger, LLC, New York, NY, 2019) using the OPLS3e force-field. This involved ionization using Epik to generate possible protonation states at target pH 7.0 $\pm$ 2.0, desalting (if applicable) and generating tautomers (if applicable). The compound was then energetically minimized, and the resulting 3-dimensional structure used to specify chiral centers (if applicable).

Protein Structure Preparation

The relevant X-ray structures of HDAC6 catalytic domain 2 (PDB 5EDU, 5WGI, 5WGL) and HDAC8 in (PDB 1T64) were retrieved from the protein data bank (https://www.rcsb.org) into Maestro 12.2.012 before undergoing a protein preparation workflow (Protein Preparation Wizard, Prime, Schrödinger, LLC, New

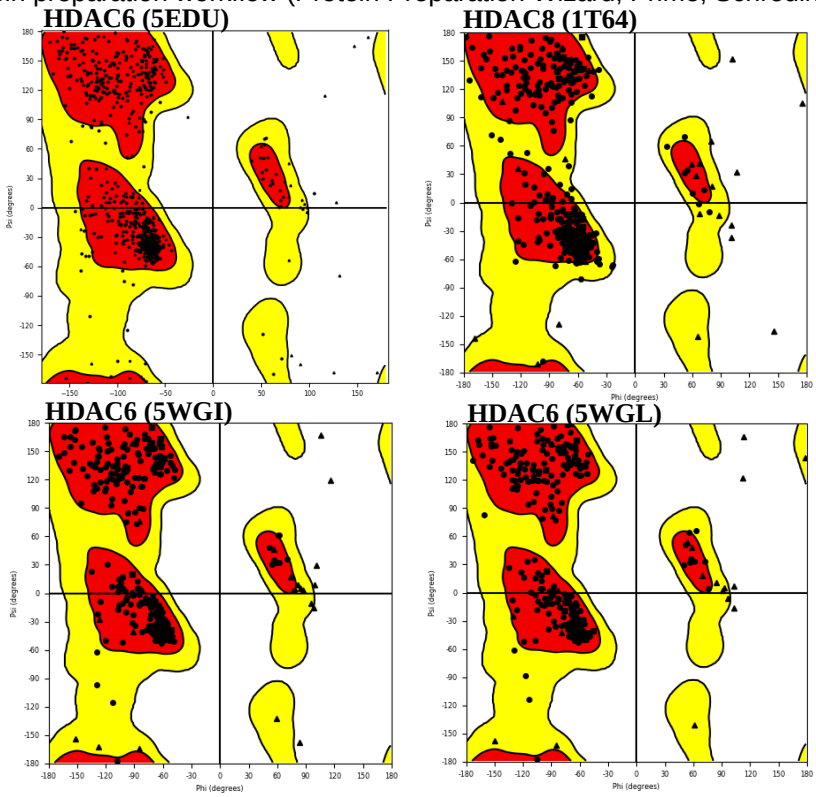


Fig. S11 Ramachandran plot outputs of HDAC6 and HDAC8 post protein-preparation. As expected, due to their structural flexibility, only Gly and Ala residues were found to occupy the disfavoured region (quadrant IV, bottom right). No other sterically forbidden angles were observed in other residues of either protein structures.

York, 2019). This procedure involved assigning appropriate bond orders, adding hydrogens, creating zero-order bonds to metal centers and the formation of disulfide bonds (where appropriate). Missing loops and residue side-chains were added using BLAST homology searches and Prime, water molecules farther than 5.0 Å from heteroatoms were deleted and, similar to the ligand preparation workflow, Epik was used to generate relevant residue ionizations at target pH 7.0 ± 2.0. All residues with reported steric clashes were individually minimized (OPLS3e). Finally, water molecules with less than 3 H-bonds to non-water molecules were removed. Using Maestro 12.2.012, Ramachandran plot analysis was used to analyze the protein structure output, ensuring successful preparation prior to *in silico* docking studies.

Receptor Grid Generation

In order to define an appropriate receptor-grid for the molecules under study, the Induced-Fit Docking (IFD) workflow by Maestro was utilized against HDAC6 **(Schrödinger Release 2019-2:** Schrödinger Suite 2019-2 Induced Fit Docking protocol; Glide, Schrödinger, LLC, New York, NY, 2016; Prime, Schrödinger, LLC, New York, NY, 2019). Using the standard protocol, with the OPLS3e FF, the grid was defined surrounding the co-crystallized ligands within the lysine tunnel (catalytic active site) of the relevant PDB for HDAC 6 and 8. This was followed by trimming neighbouring side chains (Receptor Van der Waals scaling 0.70), whilst refining residues within 5 Å of the docked ligand poses via a standard precision (SP) workflow. A metal co-ordination constraint was also applied (rewarding ligands which co-ordinate Zn^{2+}).

Ligand Docking

The relevant HDACi were screened against HDAC6 and HDAC8 using IFD protocols and the top binding poses as a function of docking score (kcal/mol) were generated and analyzed.In all docking calculations, ligands were flexible, and the protein remained static (excluding rotatable groups and the generated IFD models). The ligands were allowed to sample nitrogen inversions and ring conformations, while non-planar amide conformations were penalized, and intra-molecular hydrogen bonds were rewarded. After post-docking minimizations, the top binding pose for each ligand was produced and analyzed for key interactions using Maestro 12.2.012 and Ligand Interaction Diagrams. Raw data for the lead compound docking studies can be seen below.

Table S4: Ligand Interaction Diagrams.

Protein	Title	State Penalty	Force Field	docking score	glide gscore	glide lipo	glide hbond	glide metal	glide rewards	glide evdw
HDAC6	TO-317	2.5805	OPLS3e	-9.065	-9.584	-2.293	-0.578	-2.3	-0.399	-34.585
HDAC6	TO-130	2.5731	OPLS3e	-8.464	-8.507	-2.784	-0.32	-2.3	0.231	-31.251
HDAC6	Citarinostat	3.0499	OPLS3e	-7.939	-8.397	-1.375	-0.566	-2.3	-0.45	-31.462
HDAC8	TO-317	2.5805	OPLS3e	-7.565	-10.146	-2.489	-0.608	-2.3	-0.574	-38.912

Protein	Title	glide ecoul	glide erotb	glide esite	glide emodel	glide energy	glide einternal	glide confnum	glide posenum	glide eff state penalty
HDAC6	TO-317	-16.84	0.46	-0.219	-90.259	-51.425	10.741	125	313	0.519
HDAC6	TO-130	-13.276	0.431	-0.212	-74.203	-44.527	7.877	90	58	0.043
HDAC6	Citarinostat	-20.284	1.111	-0.2	-103.17	-51.746	7.383	879	177	0.458
HDAC8	TO-317	-16.485	0.46	-0.217	-98.324	-55.397	9.734	107	206	2.58

X-ray Experimental Procedure

HDAC6 from *Danio rerio* (zebrafish; catalytic domain 2, henceforth abbreviated simply “HDAC6”) was expressed in *Escherichia coli* and purified as described.[4] The HDAC6–TO-317 complex was co-crystallized using the sitting drop vapor diffusion method at 4 °C by equilibrating a 200 nL drop containing 100 nL of protein solution [10 mg/mL HDAC6, 50 mM 4-(2-hydroxyethyl)-1-piperazineethanesulfonic acid (HEPES) (pH 7.5), 100 mM KCl, 5% glycerol (v/v), 1 mM tris(2-carboxyethyl)phosphine (TCEP), and 2 mM TO-317] and 100 nL of precipitant solution [0.2 M potassium formate and 20% polyethylene glycol (PEG) 3350 (w/v)] against an 80 µL reservoir of precipitant solution. Crystals appeared within 4 days. Crystals were soaked in mother liquor augmented with 30% ethylene glycol prior to harvesting and flash cooling in liquid nitrogen.

X-ray diffraction data were collected at the Highly Automated Macromolecular Crystallography (AMX) beamline 17-ID-1 at the National Synchrotron Light Source II, Brookhaven National Laboratory

(Upton, NY). Crystals of the HDAC6–TO-317 complex diffracted X-rays to 1.84 Å resolution. Diffraction data were indexed and integrated using iMosflm[5] and scaled using Aimless[6] in the CCP4 program suite.[7] The crystal structure was solved using molecular replacement as implemented in Phaser;[8] the atomic coordinates of unliganded HDAC6 (PDB 5EEM)[9] were used as a search model. The program Phenix[10] was used for crystallographic refinement and the graphics program Coot[11] was used to build and manipulate the atomic model of the HDAC6–TO-317 complex. Solvent molecules and the atomic coordinates of TO-317 were built into well-defined electron density during the later stages of refinement. The final model was evaluated using MolProbity.[12] Data collection and refinement statistics are recorded in Table S5.

Table S5. Data Collection and Refinement Statistics for the HDAC6–TO-317 Complex[a]

Space group	$P2_12_12_1$
a,b,c (Å)	75.50, 96.45, 98.45
α, β, γ (°)	90.00, 90.00, 90.00
R_{merge}[b]	0.128 (0.391)
R_{pim}[c]	0.083 (0.269)
$CC_{1/2}$[d]	0.994 (0.956)
Redundancy	6.3 (5.8)
Completeness (%)	99.3 (98.6)
I/σ	9.0 (4.4)
Refinement	
Resolution (Å)	59.43–1.84 (1.90–1.84)
No. reflections	122423 (11945)
R_{work}/R_{free}[e]	0.168/0.207 (0.187/0.246)
Number of Atoms[f]	
Protein	5486
Ligand	95
Solvent	381
Average B factors (Å^2)	
Protein	11
Ligand	18
Solvent	18
RMS Deviations	
Bond lengths (Å)	0.007
Bond angles (°)	1.2
Ramachandran Plot	
Favored	97.00
Allowed	3.00
Outliers	0.00

[a]Values in parentheses refer to the highest-resolution shell indicated. [b]$R_{merge} = \sum_{hkl}\sum_i|I_{i,hkl} - \langle I\rangle_{hkl}|/\sum_{hkl}\sum_i I_{i,hkl}$, where $\langle I\rangle_{hkl}$ is the average intensity calculated for reflection *hkl* from replicate measurements. [c]$R_{p.i.m.} = (\sum_{hkl}(1/(N-1))^{1/2}\sum_i|I_{i,hkl} - \langle I\rangle_{hkl}|)/\sum_{hkl}\sum_i I_{i,hkl}$, where $\langle I\rangle_{hkl}$ is the average intensity calculated for reflection *hkl* from replicate measurements and N is the number of reflections. [d]Pearson correlation coefficient between random half-datasets. [e]$R_{work} = \sum||F_o| - |F_c||/\sum|F_o|$ for reflections contained in the working set. $|F_o|$ and $|F_c|$ are the observed and calculated structure factor amplitudes, respectively. R_{free} is calculated using the same expression for reflections contained in the test set held aside during refinement. [f]Per asymmetric unit. [g]Calculated with MolProbity.

In Vitro HDAC IC_{50} Determination

The in vitro HDAC IC_{50} determination was carried out by Nanosyn Inc., Santa Clara, CA and the details has been reported elsewhere.[13] Briefly, an activity based electrophoretic mobility shift assay (EMSA) experiment is conducted with a microfluidic electrophoresis technology (Caliper LabChip ® 3000, Caliper Life Sciences/Perkin Elmer). The deacetylation level of a FAM-labelled acetylated peptide based on compounds HDAC activity is detected via changes in the electrophoretic mobility of the peptide. HDAC proteins are diluted in an assay buffer made of 100 mM HEPES, pH 7.5, 0.1% BSA, 0.01% Triton X-100 and 25 mM KCl and 10 µL is added in a 384-well plate. Test compounds in diluted from 10 mM DMSO stock solutions are introduced using a Labcyte Echo acoustic dispensing robot. (TSA, JNJ-26481585, and MS-275 are compounds with known HDAC IC_{50} values and are used as control compounds. Other controls are a well without protein for 0% protein activity [100% acetylated peptide] and a DMSO [no inhibitor] well for 100% protein activity [0% acetylated peptide]). 10 µL FAM-labelled acetylated peptide prediluted in the assay buffer is added and the relative signal intensity from acetylated vs deacetylated peptide is measured following separation in an electrophoretic gel. The product to sum ratio (PSR) which is a concentration-dependent measure of activity is determined using the equation

$$PSR = \frac{P}{S + P}$$

Where P = product intensity, and S = substrate intensity

The % inhibition, P_{inh}, is obtained from the equation

$$P_{inh} = (PSR_{0\%inh} - PSR_{comp})/PSR_{0\%inh} - PSR_{100\%inh}) \times 100\%$$

Where PSR_{comp}, $PSR_{0\%inh}$, and $PSR_{100\%inh}$ are the PSRs in the presence of inhibitor, absence of inhibitor, and absence of protein, respectively.

IC_{50} were determined from a plot of compound concentration vs P_{inh}, fitted to a 4-parameter sigmoidal dose-response curve on the XLfit software.

To determine the inhibition kinetics of the compounds, the enzyme inhibition in 10 samples at compounds' concentrations ranging from 3.6 to 18.8 nM were monitored for a period of ~5 h. The time-dependent inhibition data were fitted using a hyperbolic fitting algorithm to determine the observed rate constants (K_{obs}) at each concentration. The K_{obs} were plotted against [compound] and a 2-step reversible inhibition model (See below) was used to determine Ki as: $K_{obs} = k_{-2} + k_2 \times [I] / ([I] + Ki)$

$$E + I \underset{k_{-1}}{\overset{K_1}{\rightleftharpoons}} E{:}I \underset{k_{-2}}{\overset{k_2}{\rightleftharpoons}} E{:}I^*$$

Determination of Covalency Using Intact Mass Analysis

The covalent modification of the purified proteins (HDACs) with the compounds were evaluated using intact mass analysis by liquid chromatography-mass spectrometry instrument (LC-MS/MS).

The reaction solution (50 µL) was prepared in a 1.5-mL Eppendorf tube and contained the protein (10 µM), the compound (50 µM, 5:1 ratio), HEPES buffer (20 mM, pH 8), 2% DMSO, 2% glycerol, and 40 mM NaCl. The reaction was allowed to proceed for 2 h at 30 ºC. Thereafter, the reaction was quenched by the addition of 200 µL of cold acetone and incubated at -20 ºC for 1 h. Then, the tube was centrifuged for 10 min at 10,000 ×g, the supernatant was discarded, and the pellet was washed by adding 200 µL of cold acetone and centrifugation at 10,000 ×g for 10 min. The pellet was re-dissolved in 50 µL of 80% formic acid and incubated at -20 ºC for 10 min. 10 µL of the solution was diluted in 90 µL of MQ water, and 0.5 µL was injected into the LC/MS/MS for intact mass analysis.

The LC-MS/MS instrument comprises of a Waters G2-XS quadrupole-time of flight (QTof) mass spectrometer and a Waters Acuity M-class Ultra-High Performance Liquid Chromatography (UPLC) system. The M-class UPLC system includes a micro binary solvent manager (µBSM), a micro sample manager (µSM), and an IonKey (iKey) separation system. The mobile phase consisted of: A) 0.1% (v/v) formic acid in MilliQ water; B) 0.1% (v/v) formic acid acetonitrile. Gradients were run over 12 min and proceeded as follows: A:B, 85:15, 0.0 – 1 min, 85:15 → 10:90, 1 – 7 min, 10:90, 7 – 9 min, 10:90 → 85:15, 9 – 9.5 min, 85:15, 9.5 – 12 min. The analytical column was a Waters BEH C4 iKey 1.7 µm (50 × 0.15 mm) column with pore sizes of 300 Å. The TOF MS data was collected in positive ion mode (m/z of 400-2000 Da) using MassLynx software (Waters). The spectral deconvolution was performed using UNIFI software (Waters). The added mass of the protein upon covalent modification to cysteine residues were specified. Multiple modifications were allowed. All the adducts with signal intensities of <2% of the base peak were ignored. The % modification was calculated as the adduct signal intensity over the total intensities of the protein peaks and the adducts.

Plasma Stability Study

The plasma stability of the compounds was evaluated in human plasma provided from Sigma Aldrich, Oakville, Canada. The main stock solutions of the compounds were prepared at 10 mM in DMSO, which further diluted to 100 µM in 50% acetonitrile (ACN) / 50% water. Aliquots of 356.5 µL of plasma proteins (2 aliquots per compound) were transferred to a test vial. The vials were placed in a heating block and were allowed to equilibrate at 37 ºC for 5 min. Then, 3.5 µL of 100 µM compound solutions were added to each vial (final concentration of 1 µM) to start the reaction. Three quality control (QC) samples at 100 nM, 500 nM, and 1000 nM were prepared in 0.5% ACN independently of those used for the plasma stability assays. The vial contents were transferred in 50 µL aliquots at time points of 0, 15, 30 and 60 minutes to a 96-well autosampler plate containing 150 µL of protein precipitation solution to quench the reactions. Ice-cold acetonitrile containing internals standards (200 nM Dexamethasone) was used as the protein precipitation solution. After centrifugation of the plate at 4 ºC, 5500 rpm for 15 min, the supernatant was diluted 10 times in MQ water. The diluted solution (5 µL) was injected into the Liquid Chromatography-Mass Spectrometry instrument (LC/MS/MS) for quantitative analysis.

The LC-MS/MS instrument comprises of a Waters G2-XS quadrupole-time of flight (QTof) mass spectrometer and a Waters Acuity I-class Ultra-High-Performance Liquid Chromatography (UPLC) system. The M-class UPLC system includes a binary solvent manager (BSM), an Acquity sample manager (SM), and a Waters T3 BEH Peptide C18 1.7 µm (50 × 2.1 mm) column. The mobile phase consisted of: A) 0.1% (v/v) formic acid in MilliQ water; B) 0.1% (v/v) formic acid acetonitrile. Gradients were run over 3 min and proceeded as follows: A:B, 85:15, 0.0 – 0.2 min, 85:15 → 10:90, 0.2 – 1 min, 10:90, 1 – 2 min, 10:90 → 85:15, 2 – 2.1 min, 85:15, 2.1 – 3 min. The MS data was collected via high resolution MS (HRMS) in positive ion mode.

All calculations were carried out using Microsoft Excel. Peak areas were determined from extracted ion chromatograms. The in vitro half-life (t1/2) of parent compounds were determined by regression analysis of the percent parent disappearance vs. time curve.

Whole Blood Stability

The whole blood stability of the compounds was evaluated in human whole blood (Pooled gender, anticoagulant: K_3EDTA) provided from BioIVT, Hicksville, New York, United States. The main stock solutions of the compounds were prepared at 10 mM in DMSO, which further diluted to 100 µM in 50% acetonitrile (ACN) / 50% water. Aliquots of 356.5 µL of plasma proteins (2 aliquots per compound) were transferred to a test vial. The vials were placed in a heating block and were allowed to equilibrate at 37 ºC for 5 min. Then, 3.5 µL of 100 µM compound solutions were added to each vial (final concentration of 1 µM) to start the reaction. Three quality control (QC) samples at 100 nM, 500 nM, and 1000 nM were prepared in 0.5% ACN independently of those used for the plasma stability assays. The vial contents were transferred in 50 µL aliquots at time points of 0, 15, 30 and 60 minutes to a 96-well autosampler plate containing 150 µL of protein precipitation solution to quench the reactions. Ice-cold acetonitrile containing internals standards (200 nM Dexamethasone) was used as the protein precipitation solution. After centrifugation of the plate at 4 ºC, 5500 rpm for 15 min, the supernatant was diluted 10 times in MQ water.

The diluted solution (5 µL) was injected into the Liquid Chromatography-Mass Spectrometry instrument (LC/MS/MS) for quantitative analysis.

The LC-MS/MS instrument comprises of a Waters G2-XS quadrupole-time of flight (QTof) mass spectrometer and a Waters Acuity I-class Ultra-High-Performance Liquid Chromatography (UPLC) system. The M-class UPLC system includes a binary solvent manager (BSM), an Acquity sample manager (SM), and a Waters T3 BEH Peptide C18 1.7 µm (50 × 2.1 mm) column. The mobile phase consisted of: A) 0.1% (v/v) formic acid in MilliQ water; B) 0.1% (v/v) formic acid acetonitrile. Gradients were run over 3 min and proceeded as follows: A:B, 85:15, 0.0 – 0.2 min, 85:15 → 10:90, 0.2 – 1 min, 10:90, 1 – 2 min, 10:90 → 85:15, 2 – 2.1 min, 85:15, 2.1 – 3 min. The MS data was collected via high resolution MS (HRMS) in positive ion mode.

All calculations were carried out using Microsoft Excel. Peak areas were determined from extracted ion chromatograms. The in vitro half-life ($t_{1/2}$) of parent compounds were determined by regression analysis of the percent parent disappearance vs. time curve.

***In Vivo* PK Study**

The *in vivo* pharmacokinetics (PK) was evaluated in Balb/C male mice. In order to prepare the dose, the test compound was dissolved in 1% DMSO, 50% PEG-400, 5% TWEEN-80, 44% Water at 10 mg/mL using the following procedure: the test compound was weighted in a clean glass vial (11.25 mg) and 0.011 mL of DMSO, 0.563 mL of PEG400, 0.056 mL of TWEEN-80, and 0.495 mL of Water were added subsequently into the glass vial. The vial was vortexed and sonicated following the addition of each reagent. to get a 10 mg/ mL solution. Three male CD-1 mice of approximately 6-8 weeks of age, and 20-30 g of weight obtained from Zhejiang Vital River Laboratory Animal Technology Co., Ltd (China) were selected for IP dosing. The mice had free access to food and water, were inspected for clinical signs and were weighted once prior to dosing. The mice were dosed with the test compound (50 mg/Kg, IP) once. The blood (30 µL) was collected at dorsal metatarsal vein (heart puncture for 12-h timepoint) at 5, 15, 30 min, 1, 2, 4, 6, 8, 12 hours post dose. The blood was transferred into a collection tube with heparin as an anticoagulant, centrifuged at 4000 g for 5 minutes at 4℃. The plasma samples were stored in a freezer at -75±15°C prior to analysis.

The desired serial concentrations of working solutions were achieved by diluting stock solution of analyte with 50% acetonitrile in MQ solution. 5 µL of working solutions (2, 4, 20, 100, 200, 1000, 2000, 4000 ng/mL) were added to 10 µL of the blank CD1 mouse plasma to achieve calibration standards of 1~1000 ng/mL (1, 2, 10, 50, 100, 500, 1000, 2000 ng/mL) in a total volume of 15 µL. Four quality control (QC) samples at 2 ng/mL, 5 ng/mL, 50 ng/mL and 1600 ng/mL for plasma were prepared independently of those used for the calibration curves. These QC samples were prepared on the day of analysis in the same way as calibration standards.

15 µL of standards, QC samples, or unknown samples（10 µL plasma with 5 µL blank solution）were added to 200 µL of acetonitrile containing IS mixture (Verapamil 2 ng/mL, Dexamethasone 50 ng/mL) for precipitating protein, respectively. Then the samples were vortexed for 30 s. After centrifugation at 4 ºC, 3900 rpm for 15 min, the supernatant was diluted 3 times with MQ water. The diluted solution (3 µL) was injected into the Liquid Chromatography-Mass Spectrometry instrument (LC/MS/MS) for quantitative analysis.

The LC-MS/MS instrument comprise of an API 5500 mass spectrometer (AB Inc. Canada) (serial# EF20351803) and a Shimadzu High Performance Liquid Chromatography (HPLC) system. The HPLC comprises of several modules including DGU-20A5R (serial# L20705518892 IX), LC-30AD (serial# L20555510778 AE and L20555510647 AE), SIL-30AC (serial# L20565504943 AE), Rack Changer II (serial# L20585501071 SS), CTO-30A (serial# L20575501294 CD), and CBM-20A (serial# L20235533959 CD). The mobile phase consisted of: A) 0.1% (v/v) formic acid in MilliQ water/acetonitrile (95:5); B) 0.1% (v/v) formic acid in MilliQ water/acetonitrile (5:95). Gradients were run over 2.5 min and proceeded as follows: A:B, 90:10, 0.0 – 0.2 min, 90:10 → 10:90, 0.2 – 1.9 min, 10:90, 1.9 – 2.2 min, 10:90 → 90:10, 2.20 – 2.21 min, 90:10, 2.21 – 2.5 min. The analytical column was an Agilent ZORBAX XDB-Phenyl 5 µm (50 × 2.1 mm) column. The MS data was collected via multiple reaction monitoring in positive ion mode.

The PK data was analyzed using WinNonlin (PhoenixTM, version 6.1). The following pharmacokinetic parameters were calculated from the plasma concentration versus time data via noncompartmental analysis method: $T_{1/2}$, C_{max}, AUC_{last}, AUC_{inf}, MRT_{inf}, Cl.

Kinetic Solubility of lead compounds in PBS by HPLC

Solid compounds were weighed fresh in a 2-mL vial and diluted with DMSO to obtain 10 mM stock solutions. To prepare a calibration curve, an aliquot of the 10 mM stock solution was diluted with 100% DMSO to five different concentrations (20 – 500 µM). The remaining aliquot of the 10 mM stock solution was diluted in three vials to 300 µM in PBS pH 7.4, 5% DMSO for kinetic solubility. The triplicate solubility sample solutions were stirred at 1000 rpm for 2 hours. Afterwards, the sample solutions were immediately filtered through a 0.45 µm nylon filter and the filtered solution was diluted 1:1 with 100% DMSO into an analytical HPLC vial. The calibration curve samples and solubility samples were injected onto an Agilent ZORBAX XDB HPLC column (C18) at 1.2 mL/min with a gradient of 98% H_2O:2% ACN (+ 0.1% formic acid) to 100% ACN (+ 0.1% formic acid) over 8 min and then 2 min at 100% ACN, 0.1% formic acid. The absorbance values at 254 nm which corresponded to each compound were integrated and a calibration curve was constructed by plotting absorbance integration vs concentration. The integration of the kinetic solubility samples was related to the slope of the calibration curve, also accounting for the final 1:1 dilution, to calculate the kinetic solubility of each compound.

PAMPA Assay

The permeability of the compounds was evaluated using the PAMPA assay. The PAMPA assembly was consisted of the acceptor plate (MultiScreen IP Filter Plate, Millipore Sigma, Canada) and the donor plate (96 well Collection Plate, Millipore Sigma, Canada). The artificial membrane solution was prepared as 1.8 % solution (w/v) of lecithin (Sigma Aldrich, Oakville, Canada) in dodecane and the mixture was sonicated to ensure a complete dissolution. Five µL of the lecithin/dodecane mixture was pipetted into each acceptor plate well (top compartment). Thereafter, 300 µL of PBS (1X PBS, pH 7.4, 5% DMSO) solution was added to each well of the acceptor plate and 300 µL of drug-containing donor solutions (50 µM compounds in 51X PBS, pH 7.4, 5% DMSO) to each well of the donor plate (bottom compartment) in triplicate. The acceptor plate was placed into the donor plate and the assembly was incubated at room temperature for 16 hours. After incubation, aliquots of 10 µL from each well of acceptor and donor plate were transferred into a 96-well plate and 190 µL of acetonitrile (containing IS: 300 nM Dexamethasone, 100 nM Phenacetin), was added into each well. The plate was vortexed at 750 rpm for 2 min and was centrifuged at 7,000 g for 10 minutes. The concentration of the compounds was determined by LC/MS/MS.

The effective permeability (Pe), in units of centimeter per second, was calculated using the following equation:

$$Log\ P_e = Log\left\{ C \times \left[-Ln\left(1 - \frac{[drug]_{acceptor}}{[drug]_{equilibrium}} \right) \right] \right\}$$

Where: C = VD × VA / [(VD + VA) × t × A]; VD = volume of donor compartment (0.30 mL); VA = volume of acceptor compartment (0.30 mL); A = filter area (0.24 cm^2 for Multi-Screen Permeability Filter plate); and t = incubation time (in seconds).

Product Characterization

Product A (Secondary Sulfonamides):

Compound 2A: Yield 68%. ^{1}H NMR (400 MHz, Chloroform-*d*) δ 7.31 – 7.22 (m, 5H), 5.41 (s, 1H), 4.40 (d, *J* = 6.3 Hz, 2H). ^{19}F NMR (377 MHz, Chloroform-*d*) δ -136.5 – -137.5 (m), -146.4 (tt, *J* = 21.0, 6.7 Hz), -158.9 (tt, *J* = 21.3, 6.5 Hz). ^{13}C NMR (101 MHz, Chloroform-*d*) δ 145.5, 145.0, 142.8 (d, *J* = 12.6 Hz), 138.8 (d, *J* = 21.3 Hz), 137.3, 134.8, 60.5, 50.8 – 49.2 (m), 49.1 (d, *J* = 4.8 Hz), 47.7, 47.5, 14.2. MS ESI+ Found 360.21 ($M+Na^+$), ESI- Found 336.17 ($M-H^+$), 372.23 ($M+Cl^-$)

Compound 3A: Yield 78%. ^{1}H NMR (400 MHz, Chloroform-*d*) δ 7.29 – 7.23 (m, 2H), 7.04 – 6.98 (m, 2H), 5.50 – 5.34 (m, 1H), 4.36 (d, *J* = 6.2 Hz, 2H). ^{19}F NMR (376 MHz, Chloroform-*d*) δ -112.9 (td, *J* = 8.6, 4.4 Hz), -136.6 – -137.0 (m), -145.9 (tt, *J* = 20.8, 6.4 Hz), -158.5 (tt, *J* = 20.9, 6.4 Hz). ^{13}C NMR (101 MHz, $CDCl_3$) δ 163.9 161.4, 130.9, 130.8, 129.9, 129.9, 129.7, 129.6, 129.4, 129.3, 116.3, 116.1, 115.9, 115.8, 115.7, 48.5, 47.0, 46.8. MS ESI- Found 354.31 ($M-H^+$)

Compound 4A: Yield 76%. ^{1}H NMR (400 MHz, Chloroform-*d*) δ 7.58 (d, *J* = 8.0 Hz, 2H), 7.43 (d, *J* = 8.0 Hz, 2H), 5.85 (t, *J* = 6.4 Hz, 1H), 4.45 (d, *J* = 6.3 Hz, 2H). ^{19}F NMR (376 MHz, Chloroform-*d*) δ -63.0, -136.3 – -137.8 (m), -145.6 (tt, *J* = 20.7, 6.5 Hz), -157.9 – -159.8 (m). ^{13}C NMR (101 MHz, Chloroform-*d*) δ 139.1, 130.9, 130.5, 128.4, 125.7, 125.7, 125.7, 125.6, 125.1, 122.4, 47.1. MS ESI- Found 404.07 ($M-H^+$)

Compound 5A: Yield 53%. ^{1}H NMR (400 MHz, Chloroform-*d*) δ 7.10 – 6.98 (m, 2H), 6.59 – 6.48 (m, 2H), 5.47 (t, *J* = 6.3 Hz, 1H), 4.30 (d, *J* = 6.2 Hz, 2H), 2.93 (s, 6H). ^{19}F NMR (376 MHz, Chloroform-*d*) δ -135.0 – -138.7 (m), -147.8 (tt, *J* = 21.2, 6.2 Hz), -158.7 – -162.4 (m). ^{13}C NMR (101 MHz, $CDCl_3$) δ 150.3, 142.7, 136.2, 129.3, 122.0, 117.2, 111.9, 47.7, 40.3. MS ESI- Found 379.32 ($M-H^+$)

Compound 6A: ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.24 (s, 2H), 7.10 (d, *J* = 4.6 Hz, 2H), 5.44 (t, *J* = 6.3 Hz, 1H), 4.35 (d, *J* = 6.3 Hz, 2H), 2.33 (s, 3H). ^{19}F NMR (376 MHz, $CDCl_3$) δ -136.3 – -137.4 (m), -147.0 (tt, *J* = 20.8, 6.4 Hz), -158.7 – -160.0 (m). ^{13}C NMR (101 MHz, $CDCl_3$) δ 138.5, 138.4, 134.1, 131.6, 129.9, 129.5, 129.3, 128.1, 127.8, 127.7, 127.6, 49.1, 49.0, 47.7, 34.7, 25.3, 21.2, 21.1, 21.0. MS ESI Found 350.20 ($M-H^+$), 386.27 ($M+Cl^-$); ESI+ Found 374.25 ($M+Na^+$).

Compound 7A: Yield: 86%. ^{1}H NMR (400 MHz, Chloroform-*d*) δ 7.31 – 7.26 (m, 2H), 7.15 (d, *J* = 8.1 Hz, 2H), 5.44 (t, *J* = 6.4 Hz, 1H), 4.38 (d, *J* = 6.4 Hz, 2H), 1.29 (s, 9H). ^{19}F NMR (376 MHz, Chloroform-*d*) δ -136.5 – -137.0 (m), -146.5 (tt, *J* = 20.8, 6.3 Hz), -159.2 (tt, *J* = 21.3, 6.4 Hz). ^{13}C NMR (101 MHz, $CDCl_3$) δ 151.6, 145.4, 142.7, 138.8, 131.7, 128.0, 125.4, 117.1, 47.4, 34.5, 31.3, 31.1. MS ESI- Found 392.37 ($M-H^+$) and 428.34 ($M+Cl^-$). MS ESI+ found 416.31 ($M+Na^+$).

Compound 8A: ^{1}H NMR (400 MHz, $CDCl_3$) δ 8.60 – 8.49 (m, 2H), 7.32 – 7.22 (m, 2H), 6.64 (s, 1H), 4.42 (s, 2H). ^{19}F NMR (376 MHz, $CDCl_3$) δ -136.5 – -137.1 (m), -145.3 (tt, *J* = 21.1, 6.5 Hz), -158.2 (tt, *J* = 20.8, 6.4 Hz). ^{13}C NMR (101 MHz, $CDCl_3$) δ 149.8, 145.3, 122.5, 60.4, 46.2, 21.1, 14.2. MS ESI- Found 337.18 ($M-H^+$), ESI+ Found 339.23 ($M+H^+$).

Compound 9A: Yield: 67%. ^{1}H NMR (400 MHz, Acetonitrile-d_3) δ 8.57 – 8.46 (m, 2H), 7.59 (dddd, *J* = 9.5, 8.2, 6.0, 2.5 Hz, 1H), 7.29 – 7.21 (m, 2H), 6.92 (s, 1H), 4.25 (s, 2H). ^{19}F NMR (376 MHz, Acetonitrile-d_3) δ -136.5 – -136.8 (m), -138.5 – -138.8 (m), -149.5 – -150.2 (m), -154.0 – -154.6 (m). ^{13}C NMR (101 MHz, Acetonitrile-d_3) δ 206.5, 149.8, 149.7, 145.9, 122.4, 112.2, 112.2, 112.0, 112.0, 45.3, 29.9. MS ESI- Found 319.13 ($M-H^+$), ESI+ Found 321.27 ($M+H^+$)

Compound 10A: ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.47 (tdd, *J* = 8.5, 5.8, 2.5 Hz, 1H), 7.28 (dt, *J* = 5.2, 2.5 Hz, 3H), 7.20 (dd, *J* = 7.1, 2.6 Hz, 2H), 5.42 (t, *J* = 6.2 Hz, 1H), 4.28 (d, *J* = 6.2 Hz, 2H). ^{19}F NMR (376 MHz, $CDCl_3$) δ -135.2 (ddt, *J* = 21.3, 13.9, 7.0 Hz), -135.6 – -136.3 (m), -146.7 (tt, *J* = 20.0, 8.1 Hz), -151.6 (t, *J* = 20.2 Hz). ^{13}C NMR (101 MHz, $CDCl_3$) δ 147.6, 143.2, 142.2, 135.2, 128.7, 128.3, 128.0, 125.4, 112.1, 112.1, 111.9, 111.8, 47.5. MS ESI+ Found 320.19 ($M + H^+$), 342.33 ($M + Na^+$), 661.25 ($2M + Na^+$); ESI- Found 318.23 ($M - H^+$), 351.20 ($M + Cl^-$), 637.49 ($2M – H^+$)

Compound 11A: Yield: 88%. ^{1}H NMR (400 MHz, Chloroform-*d*) δ 7.51 (dddd, *J* = 8.6, 7.7, 6.1, 2.5 Hz, 1H), 7.25 – 7.18 (m, 2H), 7.04 – 6.95 (m, 2H), 5.39 (t, *J* = 6.2 Hz, 1H), 4.25 (d, *J* = 6.2 Hz, 2H). ^{19}F NMR

(376 MHz, Chloroform-*d*) δ -112.9 – -113.9 (m), -135.2 – -135.6 (m), -146.2 (tt, *J* = 19.0, 7.9 Hz), -151.0 – -151.7 (m). ^{13}C NMR (101 MHz, Chloroform-*d*) δ 163.8, 161.3, 131.2, 131.2, 129.8, 129.7, 115.8, 115.6, 112.1, 112.0, 111.9, 111.8, 46.7. MS ESI- Found 336.15 and 372.24 (M + Cl^-), ESI+ Found 360.23 (M + Na^+)

Compound 12A: Yield: 77%. ^{1}H NMR (400 MHz, Acetonitrile-d_3) δ 7.55 (dddd, *J* = 9.6, 8.3, 6.0, 2.5 Hz, 1H), 7.36 – 7.26 (m, 1H), 7.10 – 7.05 (m, 1H), 7.05 – 6.97 (m, 2H), 6.67 (s, 1H), 4.23 (s, 2H). ^{19}F NMR (376 MHz, Acetonitrile-d_3) δ -114.9 (td, *J* = 9.4, 5.9 Hz), -136.4 – -137.4 (m), -138.5 – -139.5 (m), -150.1 (tt, *J* = 19.1, 8.0 Hz), -154.3 – -154.9 (m). ^{13}C NMR (101 MHz, Acetonitrile-d_3) δ 163.8, 139.5, 139.4, 130.4, 130.3, 123.8, 123.8, 114.7, 114.5, 114.4, 114.2, 112.2, 112.2, 112.0, 112.0, 46.0, 46.0. MS ESI- Found 336.15

Compound 13A: Yield: 79%. ^{1}H NMR (400 MHz, Acetonitrile-d_3) δ 7.50 (dddd, *J* = 9.6, 8.3, 6.0, 2.5 Hz, 1H), 7.38 – 7.23 (m, 2H), 7.12 (td, *J* = 7.5, 1.2 Hz, 1H), 7.07 – 6.96 (m, 1H), 6.63 (s, 1H), 4.30 (d, *J* = 6.0 Hz, 2H). ^{19}F NMR (376 MHz, Acetonitrile-d_3) δ -119.7 (dt, *J* = 12.2, 6.5 Hz), -137.0 (ddt, *J* = 20.4, 13.3, 7.1 Hz), -138.7 – -139.2 (m), -150.1 (ddd, *J* = 27.4, 18.6, 8.1 Hz), -154.4 – -155.0 (m). ^{13}C NMR (101 MHz, Acetonitrile-d_3) δ 161.8, 159.4, 130.7, 130.7, 130.1, 130.0, 124.3, 124.3, 123.4, 123.3, 115.2, 114.9, 112.2, 112.1, 112.0, 111.9, 40.5, 40.5. MS ESI- Found 336.17

Compound 14A: Yield: 85%. ^{1}H NMR (400 MHz, DMSO-d_6) δ 9.02 (s, 1H), 8.39 (dt, *J* = 4.9, 1.3 Hz, 1H), 7.74 (td, *J* = 7.7, 1.8 Hz, 1H), 7.66 (dddd, *J* = 10.1, 8.2, 6.0, 2.5 Hz, 1H), 7.34 (d, *J* = 7.8 Hz, 1H), 7.24 (ddd, *J* = 7.6, 4.8, 1.1 Hz, 1H), 4.28 (s, 2H). ^{19}F NMR (376 MHz, DMSO-d_6) δ -135.3 (ddt, *J* = 26.8, 13.2, 6.9 Hz), -137.4 – -137.9 (m), -148.9 (tt, *J* = 21.6, 8.0 Hz), -153.2 (t, *J* = 21.6 Hz). ^{13}C NMR (101 MHz, DMSO-d_6) δ 156.9, 149.2, 144.7, 143.2, 137.2, 123.0, 122.3, 112.4, 112.3, 112.1, 112.1, 48.2. MS ESI- Found 319.13 ESI+ Found 321.21 and 343.19 (M + Na^+)

TO-317-A: Yield: 89%. ^{1}H NMR (400 MHz, Acetone-d_6) δ 8.49 (d, *J* = 2.3 Hz, 1H), 8.46 (dd, *J* = 4.8, 1.6 Hz, 1H), 7.93 (t, *J* = 5.8 Hz, 1H), 7.74 (dt, *J* = 7.9, 2.0 Hz, 1H), 7.63 (dddd, *J* = 10.1, 8.3, 5.9, 2.5 Hz, 1H), 7.31 (dd, *J* = 7.9, 4.8 Hz, 1H), 4.39 (d, *J* = 5.2 Hz, 2H). ^{19}F NMR (376 MHz, Acetone-d_6) δ -136.7 (ddt, *J* = 18.8, 13.3, 7.3 Hz), -138.3 – -139.0 (m), -150.4 (tt, *J* = 19.9, 8.1 Hz), -154.4 – -154.9 (m). ^{13}C NMR (101 MHz, Acetone-d_6) δ 205.4, 149.4, 148.9, 135.5, 132.6, 123.3, 112.2, 112.1, 111.9, 111.9, 44.2. MS ESI- Found 319.06 (M - H^+); ESI+ Found 320.91 (M + H^+)

<u>Product B (Tertiary Sulfonamide, Ester Derivatives):</u>

Compound 2B: Yield: 66%. ^{1}H NMR (400 MHz, Chloroform-*d*) δ 7.95 – 7.89 (m, 2H), 7.31 – 7.21 (m, 5H), 7.15 (dd, *J* = 6.6, 3.0 Hz, 2H), 4.60 (s, 2H), 4.52 (s, 2H), 1.62 (s, 9H). ^{19}F NMR (376 MHz, Chloroform-*d*) δ -134.8 – -135.3 (m), -146.4 (tt, *J* = 21.0, 6.6 Hz), -158.7 – -159.3 (m). ^{13}C NMR (101 MHz, Chloroform-*d*) δ 165.2, 139.3, 134.2, 132.0, 129.8, 128.7, 128.6, 128.4, 128.2, 81.3, 51.8, 51.5, 28.1. MS ESI- Found 562.42 (M + Cl^-), ESI+ Found 550.41 (M + Na^+), 1077.66 (2M + Na^+).

Compound 3B: Yield: 72%. ^{1}H NMR (400 MHz, Chloroform-*d*) δ 7.95 – 7.85 (m, 2H), 7.26 – 7.18 (m, 2H), 7.18 – 7.10 (m, 2H), 6.98 (dt, *J* = 8.1, 5.4 Hz, 2H), 4.56 (d, *J* = 3.6 Hz, 2H), 4.51 (d, *J* = 3.8 Hz, 2H), 1.63 (d, *J* = 3.7 Hz, 9H). ^{19}F NMR (376 MHz, Chloroform-*d*) δ -113.2 (q, *J* = 7.9 Hz), -134.4 – -136.0 (m), -145.8 (td, *J* = 21.2, 10.6 Hz), -158.6 (t, *J* = 19.8 Hz). ^{13}C NMR (101 MHz, Chloroform-*d*) δ 165.1, 161.4, 139.0, 132.1, 130.4, 130.4, 130.2, 130.2, 129.8, 128.2, 115.8, 115.6, 81.4, 51.4, 51.3, 28.2. MS ESI- Found 544.46 (M-H^+) and 580.48 (M+Cl^-); ESI+ Found 546.54 (M+H^+)

Compound 4B: Yield: 80%. ^{1}H NMR (400 MHz, Chloroform-*d*) δ 7.90 – 7.82 (m, 2H), 7.51 (d, *J* = 8.0 Hz, 2H), 7.30 (d, *J* = 8.0 Hz, 2H), 7.18 (d, *J* = 8.2 Hz, 2H), 4.58 (d, *J* = 4.7 Hz, 4H), 1.61 (s, 9H). ^{19}F NMR (376 MHz, Chloroform-*d*) δ -62.8, -134.4 – -136.6 (m), -145.5 (tt, *J* = 21.0, 6.6 Hz), -157.6 – -160.2 (m). ^{13}C NMR (101 MHz, Chloroform-*d*) δ 165.0, 138.7, 138.7, 132.1, 129.7, 128.8, 128.3, 125.6, 125.5, 81.4, 52.1, 51.8, 28.1. MS ESI Found 630.47 (M + Cl^-); ESI+ Found 618.51 (M + Na^+).

Compound 5B: Yield: 55%. ^{1}H NMR (400 MHz, Chloroform-*d*) δ 8.01 – 7.90 (m, 2H), 7.37 – 7.27 (m, 2H), 7.13 – 7.04 (m, 2H), 6.99 – 6.91 (m, 2H), 4.45 (s, 2H), 4.38 (s, 2H), 2.96 (s, 6H), 1.62 (s, 9H). ^{19}F NMR (376 MHz, Chloroform-*d*) δ -134.9 (dt, *J* = 21.4, 5.6 Hz), -147.3, -159.1 – -159.6 (m). ^{13}C NMR (101

MHz, $CDCl_3$) δ 165.3, 150.4, 140.0, 131.8, 129.94, 129.9, 129.8, 128.1, 120.9, 120.7, 112.0, 111.5, 81.2, 51.5, 51.3, 40.9, 40.3, 40.2, 28.2. MS ESI- Found 605.53 ($M + Cl^-$); ESI+ Found 593.63 ($M + Na^+$).

Compound 6B: Yield: 54%. ^1H NMR (400 MHz, $CDCl_3$) δ 7.96 – 7.89 (m, 2H), 7.30 – 7.22 (m, 2H), 7.08 (d, *J* = 7.9 Hz, 2H), 7.02 (d, *J* = 8.1 Hz, 2H), 4.60 (s, 2H), 4.47 (s, 2H), 2.33 (s, 3H), 1.63 (s, 9H). ^{19}F NMR (376 MHz, $CDCl_3$) δ -135.0 (qd, *J* = 13.1, 7.3 Hz), -146.7 (tt, *J* = 21.0, 6.5 Hz), -158.9 – -159.4 (m). ^{13}C NMR (101 MHz, $CDCl_3$) δ 165.2, 145.6, 144.9, 143.1, 142.3, 139.5, 138.3, 136.4, 131.9, 131.0, 129.8, 129.33, 128.6, 128.1, 117.5, 81.3, 51.5, 51.4, 51.4, 51.4. MS ESI- Found 576.49 ($M+Cl^-$), ESI+ Found 564.52 ($M+Na^+$).

Compound 7B: Yield: 57%. ^1H NMR (400 MHz, $CDCl_3$) δ 7.92 (dd, *J* = 8.5, 2.9 Hz, 2H), 7.26 (ddd, *J* = 19.1, 8.3, 3.0 Hz, 4H), 7.11 – 7.01 (m, 2H), 4.66 (d, *J* = 3.1 Hz, 2H), 4.46 (d, *J* = 3.0 Hz, 2H), 1.63 (s, 9H), 1.29 (d, *J* = 3.0 Hz, 9H). ^{19}F NMR (376 MHz, $CDCl_3$) δ -135.1 (dq, *J* = 19.3, 6.4 Hz), -146.3 – -147.0 (m), -159.1 (t, *J* = 19.5 Hz). ^{13}C NMR (101 MHz, $CDCl_3$) δ 165.2, 151.6, 139.7, 131.8, 131.0, 129.7, 128.5, 128.1, 125.5, 81.2, 52.1, 51.6, 34.5, 31.2, 28.2. MS ESI- Found 618.54 ($M+Cl^-$); ESI+ Found 606.58 ($M+Na^+$).

Compound 8B: Yield: 62%. ^1H NMR (400 MHz, $CDCl_3$) δ 8.52 (s, 2H), 7.89 – 7.80 (m, 2H), 7.16 (d, *J* = 8.2 Hz, 2H), 7.10 (d, *J* = 4.9 Hz, 2H), 4.53 (s, 4H), 1.59 (s, 9H). ^{19}F NMR (376 MHz, $CDCl_3$) δ -134.8 – -135.3 (m), -145.1 (tt, *J* = 21.1, 6.6 Hz), -157.9 – -158.6 (m). ^{13}C NMR (101 MHz, $CDCl_3$) δ 164.9, 150.2, 145.7, 143.9, 138.2, 132.3, 129.8, 128.4, 122.8, 81.5, 51.9, 51.2, 28.1. MS ESI- Found 563.47 ($M+Cl^-$); ESI+ Found 529.46 ($M+H^+$), 551.39 ($M+Na^+$).

Compound 9B: Yield: 66%. ^1H NMR (400 MHz, Acetonitrile-d_3) δ 8.47 – 8.42 (m, 2H), 7.89 – 7.74 (m, 2H), 7.60 (dddd, *J* = 9.7, 8.2, 5.9, 2.5 Hz, 1H), 7.29 – 7.22 (m, 2H), 7.17 – 7.11 (m, 2H), 4.54 (s, 4H), 1.58 (s, 9H). ^{19}F NMR (376 MHz, Acetonitrile-d_3) δ -134.8 – -135.3 (m), -137.9 – -138.6 (m), -148.9 (tt, *J* = 19.8, 8.8 Hz), -153.4 – -153.9 (m). ^{13}C NMR (101 MHz, Acetonitrile-d_3) δ 149.8, 145.0, 140.0, 131.8, 129.2, 128.7, 122.9, 112.9, 112.6, 81.0, 52.1, 52.1, 51.3, 51.3, 27.3. MS ESI- Found 545.39 ($M+Cl^-$); ESI+ Found 511.45 ($M+H^+$) and 533.21 ($M+Na^+$)

Compound 10B: Yield: 70%. ^1H NMR (400 MHz, Acetonitrile-d_3) δ 7.88 – 7.81 (m, 2H), 7.54 (dddd, *J* = 9.8, 8.3, 5.9, 2.5 Hz, 1H), 7.31 – 7.22 (m, 5H), 7.22 – 7.15 (m, 2H), 4.54 (s, 2H), 4.51 (s, 2H), 1.59 (s, 9H). ^{19}F NMR (376 MHz, Acetonitrile-d_3) δ -135.0 – -135.8 (m), -138.1 – -139.1 (m), -149.5 (tt, *J* = 19.7, 8.6 Hz), -153.5 – -154.5 (m). ^{13}C NMR (101 MHz, Acetonitrile-d_3) δ 140.6, 135.4, 131.5, 129.2, 128.6, 128.5, 128.3, 127.9, 80.9, 52.0, 51.4, 51.4, 27.3. MS ESI+ Found 532.41 ($M + Na^+$); ESI- Found 544.38 ($M + Cl^-$)

Compound 11B: Yield: 71%. ^1H NMR (400 MHz, Chloroform-*d*) δ 7.94 – 7.87 (m, 2H), 7.56 – 7.46 (m, 1H), 7.15 (d, *J* = 1.8 Hz, 2H), 7.13 – 7.05 (m, 2H), 7.02 – 6.90 (m, 2H), 4.48 (s, 2H), 4.44 (s, 2H), 1.62 (s, 10H). ^{19}F NMR (376 MHz, Chloroform-*d*) δ -113.4 (td, *J* = 8.2, 7.6, 4.2 Hz), -135.2 – -136.1 (m), -146.2 (tt, *J* = 20.4, 8.2 Hz), -151.1 (td, *J* = 22.1, 20.6, 3.8 Hz). ^{13}C NMR (101 MHz, Chloroform-*d*) δ 165.1, 139.3, 132.0, 130.4, 130.3, 129.8, 128.1, 115.8, 115.6, 115.5, 112.5, 112.4, 112.2, 112.2, 81.3, 50.9, 50.8, 50.8, 50.7, 49.4, 28.1. MS ESI- Found 562.44 ($M + Cl^-$).; ESI+ Found 550.48 ($M + H^+$)., 1077.86 ($2M + H^+$).

Compound 12B: Yield: 68%. ^1H NMR (400 MHz, Acetonitrile-d_3) δ 7.88 – 7.76 (m, 2H), 7.57 (dddd, *J* = 9.7, 8.2, 5.9, 2.5 Hz, 1H), 7.34 – 7.19 (m, 3H), 7.05 – 6.95 (m, 2H), 6.92 (dt, *J* = 10.1, 2.1 Hz, 1H), 4.54 (s, 2H), 4.51 (s, 2H), 1.59 (s, 9H). ^{19}F NMR (376 MHz, Acetonitrile-d_3) δ -114.8 (td, *J* = 9.5, 6.0 Hz), -135.2 (ddt, *J* = 21.4, 14.0, 6.1 Hz), -138.0 – -139.0 (m), -149.2 (tt, *J* = 19.2, 8.4 Hz), -153.6 – -154.3 (m). ^{13}C NMR (101 MHz, Acetonitrile-d_3) δ 140.4, 130.4, 129.2, 128.4, 124.4, 117.3, 115.2, 115.0, 114.61, 114.4, 112.8, 112.6, 80.9, 51.8, 51.7, 27.3. MS ESI- Found 562.63 ($M + Cl^-$); ESI+ Found 550.51 ($M + Na^+$)

Compound 13B: Yield: 68%. ^1H NMR (400 MHz, Acetonitrile-d_3) δ 7.88 – 7.80 (m, 2H), 7.51 (dddd, *J* = 9.8, 8.1, 5.8, 2.5 Hz, 1H), 7.33 – 7.22 (m, 4H), 7.09 (td, *J* = 7.5, 1.2 Hz, 1H), 6.96 (ddd, *J* = 10.3, 8.7, 1.1 Hz, 1H), 4.60 (s, 2H), 4.55 (s, 2H), 1.59 (s, 9H). ^{19}F NMR (376 MHz, Acetonitrile-d_3) δ -118.6 (dt, *J* = 12.0, 6.1 Hz), -135.4 (ddt, *J* = 20.9, 13.7, 6.9 Hz), -138.0 – -139.1 (m), -148.9 – -150.2 (m), -153.8 – -154.4 (m). ^{13}C NMR (101 MHz, Acetonitrile-d_3) δ 140.8, 131.5, 131.3, 131.3, 130.3, 130.2, 129.2, 128.0, 124.4,

124.3, 115.3, 115.0, 112.7, 112.5, 80.9, 52.0, 46.1, 27.3. MS ESI- Found 562.58 ($M + Cl^-$); ESI+ Found 550.51 ($M + Na^+$)

Compound 14B: Yield: 62%. ^{1}H NMR (400 MHz, DMSO-d_6) δ 8.37 – 8.28 (m, 1H), 7.83 (d, J = 8.2 Hz, 2H), 7.78 – 7.72 (m, 1H), 7.70 (td, J = 7.7, 1.8 Hz, 1H), 7.35 (d, J = 8.1 Hz, 2H), 7.27 – 7.16 (m, 2H), 4.67 (s, 2H), 4.56 (s, 2H), 1.55 (s, 9H). ^{19}F NMR (376 MHz, DMSO-d_6) δ -133.7 (ddt, J = 20.8, 13.6, 6.8 Hz), -137.0 – -138.0 (m), -148.1 (tt, J = 21.7, 8.1 Hz), -152.9 – -153.6 (m). ^{13}C NMR (101 MHz, DMSO-d_6) δ 165.1, 162.8, 155.4, 149.4, 141.3, 137.3, 131.2, 129.6, 128.7, 123.2, 123.2, 113.2, 112.9, 81.2, 52.5, 51.9, 36.3, 31.3, 28.2, 14.6. MS ESI- Found 545.40 ($M + Cl^-$); ESI+ Found 511.44 ($M + H^+$) and 533.26 ($M + Na^+$)

TO-317-B: Yield: 60%. ^{1}H NMR (400 MHz, $CDCl_3$) δ 8.51 (dd, J = 4.8, 1.6 Hz, 1H), 8.35 – 8.28 (m, 1H), 7.93 – 7.84 (m, 2H), 7.58 – 7.47 (m, 2H), 7.22 (ddd, J = 7.9, 4.8, 0.9 Hz, 1H), 7.19 – 7.10 (m, 2H), 4.48 (s, 4H), 1.60 (s, 9H). ^{19}F NMR (376 MHz, $CDCl_3$) δ -133.5 (ddt, J = 21.7, 14.0, 7.2 Hz), -135.1 (dddd, J = 21.9, 13.0, 9.0, 3.9 Hz), -145.6 (tt, J = 20.0, 8.1 Hz), -150.7 (td, J = 22.0, 20.7, 3.7 Hz). ^{13}C NMR (101 MHz, $CDCl_3$) δ 165.0, 149.6, 145.1, 140.0, 138.9, 136.3, 132.1, 130.6, 129.9, 128.2, 123.6, 112.5, 112.5, 112.3, 112.3, 81.4, 60.4, 51.3, 51.3, 49.2, 49.1, 28.1, 21.0, 14.2. ESI- Found 545.49 (ESI + Cl^-); ESI+ Found 511.34 (ESI + H^+), 533.38 (ESI + Na^+) and 1043.67 ($2M + Na^+$).

<u>Product C: (Tertiary Sulfonamides, Acid Derivatives):</u>

Compound 2C: Yield: 79%. ^{1}H NMR (400 MHz, DMSO) δ 7.85 – 7.79 (m, 2H), 7.34 – 7.27 (m, 5H), 7.23 – 7.22 (m, 2H), 4.63 (s, 2H), 4.56 (s, 2H). ^{19}F NMR (376 MHz, Chloroform-d) δ -134.8 – -135.3 (m), -146.4 (tt, J = 21.0, 6.6 Hz), -158.7 – -159.3 (m). ^{13}C NMR (101 MHz, DMSO) δ 73.7, 73.7, 49.5, 47.2, 41.7, 36.9, 36.2, 36.1, 36.0, 35.8, 35.6, 35.4, 35.2, 35.1, 35.0, 34.9, 34.8, 34.6. MS ESI- Found 470.38 (M-H+), 506.34 ($M+Cl^-$) and 941.60 ($2M-H^+$) ESI+ Found 494.18 ($M+Na^+$)

Compound 3C: Yield: 88%. ^{1}H NMR (400 MHz, DMSO-d_6) δ 12.94 (s, 1H), 7.84 – 7.74 (m, 2H), 7.33 – 7.26 (m, 2H), 7.26 – 7.18 (m, 2H), 7.07 (t, J = 8.9 Hz, 2H), 4.61 (s, 2H), 4.55 (s, 2H). ^{19}F NMR (376 MHz, DMSO-d_6) δ -114.2 – -115.0 (m), -136.4 (d, J = 24.9 Hz), -147.2 (d, J = 22.9 Hz), -159.5 – -160.0 (m). ^{13}C NMR (101 MHz, DMSO) δ 167.4, 163.4, 161.0, 141.0, 132.9, 131.2, 131.1, 130.5, 129.7, 128.7, 115.7, 115.5, 72.7, 71.0, 66.8, 60.7, 52.3, 52.2, 44.1, 31.1. MS ESI- Found 488.24 ($M - H^+$), 524.30 ($M + Cl^-$).

Compound 4C: Yield: 89%. ^{1}H NMR (400 MHz, Acetonitrile-d_3) δ 7.91 – 7.81 (m, 2H), 7.52 (d, J = 8.0 Hz, 2H), 7.36 (d, J = 8.0 Hz, 2H), 7.29 (d, J = 8.1 Hz, 2H), 4.64 (d, J = 4.7 Hz, 4H). ^{19}F NMR (376 MHz, Acetonitrile-d_3) δ -63.2, -134.1 – -142.3 (m), -148.6 (tt, J = 20.3, 6.9 Hz), -158.7 – -165.1 (m). ^{13}C NMR (101 MHz, Acetonitrile-d_3) δ 140.7, 140.0, 129.7, 129.5, 129.2, 128.6, 125.2, 125.2, 125.2, 52.6, 52.2. MS ESI- Found 538.32 ($M - H^+$)

Compound 5C: Yield: 63%. ^{1}H NMR (400 MHz, DMSO) δ 7.90 (d, J = 7.8 Hz, 2H), 7.38 (d, J = 7.9 Hz, 2H), 7.23 – 6.90 (m, 2H), 6.65 (s, 2H), 4.68 (s, 2H), 4.44 (s, 2H), 2.99 (s, 6H). ^{19}F NMR (376 MHz, DMSO) δ -136.3 (d, J = 24.5 Hz), -148.0, -160.1 (t, J = 23.5 Hz). ^{13}C NMR (101 MHz, DMSO) δ 167.6, 150.2, 145.6, 143.0, 141.6, 139.2, 136.7, 130.6, 130.4, 129.9, 128.6, 122.9, 117.0, 113.0, 52.4, 52.3, 40.9, 30.0, 29.6. MS ESI+ Found 515.44 ($M + H^+$).

Compound 6C: Yield: 86%. ^{1}H NMR (400 MHz, DMSO) δ 12.96 (s, 1H), 7.88 – 7.80 (m, 2H), 7.35 – 7.29 (m, 2H), 7.08 (q, J = 8.0 Hz, 4H), 4.62 (s, 2H), 4.50 (s, 2H), 2.24 (s, 3H). ^{19}F NMR (376 MHz, DMSO) δ -135.8 – -136.7 (m), -147.6 (tt, J = 22.5, 6.4 Hz), -159.6 – -160.3 (m). ^{13}C NMR (101 MHz, DMSO) δ 206.9, 167.4, 141.1, 137.7, 132.3, 130.5, 129.8, 129.3, 129.0, 128.6, 72.7, 71.0, 66.8, 60.7, 52.5, 52.2, 44.1, 31.2, 21.0. MS ESI- Found 484.36 (M - H+), 520.38 ($M + Cl^-$) and 969.50 ($2M - H^+$); ESI+ Found 486.19 ($M + H^+$)

Compound 7C: Yield: 91%. ^{1}H NMR (400 MHz, DMSO) δ 12.91 (s, 1H), 7.82 (d, J = 7.8 Hz, 2H), 7.32 (d, J = 7.8 Hz, 2H), 7.20 (d, J = 8.0 Hz, 2H), 7.11 (d, J = 7.9 Hz, 2H), 4.68 (s, 2H), 4.48 (s, 2H), 1.20 (s, 9H). ^{19}F NMR (376 MHz, DMSO) δ -136.1 – -136.6 (m), -147.5 (tt, J = 22.4, 6.3 Hz), -159.9 (dq, J = 23.7, 10.7 Hz). ^{13}C NMR (101 MHz, DMSO) δ 167.4, 150.9, 141.3, 132.1, 130.5, 129.8, 128.9, 128.6, 125.4, 71.0,

52.7, 52.4, 34.6, 31.4. MS ESI- Found 526.52 (M - H+), 562.35 (M + Cl^-) and 1053.74 (2M - H^+); ESI+ Found 550.45 (M + Na^+)

Compound 8C: Yield: 80%. ^{1}H NMR (400 MHz, DMSO) δ 8.76 – 8.53 (m, 2H), 7.91 – 7.72 (m, 2H), 7.50 (q, *J* = 2.8 Hz, 2H), 7.36 (d, *J* = 8.0 Hz, 2H), 4.81 (s, 2H), 4.67 (s, 2H). ^{19}F NMR (376 MHz, DMSO) δ -136.1 (dq, *J* = 18.3, 7.4, 6.2 Hz), -146.6 (tt, *J* = 22.0, 6.2 Hz), -159.1 – -159.9 (m). ^{13}C NMR (101 MHz, DMSO) δ 167.3, 151.6, 145.9, 140.1, 130.9, 129.8, 129.5, 124.6, 53.1, 52.2, 46.2. MS ESI- Found 471.29 (M - H+), 507.25 (M + Cl^-) and 943.57 (2M - H^+); ESI+ Found 473.28 (M + H^+) 495.31 (M + Na^+)

Compound 9C: Yield: 77%. ^{1}H NMR (400 MHz, Acetonitrile-d_3) δ 9.57 (s, 5H), 8.61 – 8.42 (m, 2H), 7.89 – 7.78 (m, 2H), 7.79 – 7.64 (m, 3H), 7.37 – 7.26 (m, 2H), 4.78 (s, 2H), 4.58 (s, 2H). ^{19}F NMR (376 MHz, Acetonitrile-d_3) δ -134.5 (ddt, *J* = 21.5, 12.7, 7.4 Hz), -137.6 – -138.1 (m), -148.0 (ddd, *J* = 27.5, 18.1, 8.5 Hz), -153.0 – -153.5 (m). ^{13}C NMR (101 MHz, Acetonitrile-d_3) δ 149.1, 144.5, 140.1, 131.6, 128.8, 128.6, 122.8, 112.9, 112.9, 80.8, 52.0, 52.7, 51.0, 51.0. MS ESI- Found 453.25 (M + H^+) and 489.21 (M + Cl^-); ESI+ Found 455.27 (M + H^+) and 477.23 (M + Na^+)

Compound 10C: Yield: 81%. ^{1}H NMR (400 MHz, DMSO) δ 12.94 (s, 1H), 7.85 – 7.78 (m, 2H), 7.31 – 7.19 (m, 6H), 7.17 (dd, *J* = 7.4, 2.2 Hz, 2H), 4.56 (s, 2H), 4.50 (s, 2H). ^{19}F NMR (376 MHz, DMSO) δ -134.5 (ddd, *J* = 29.8, 13.1, 6.8 Hz), -136.6 – -137.1 (m), -147.7 (td, *J* = 21.8, 10.6 Hz), -152.1 – -152.6 (m). ^{13}C NMR (101 MHz, DMSO) δ 167.4, 141.3, 135.8, 135.5, 130.4, 129.7, 128.9, 128.8, 128.7, 128.65, 128.6, 128.2, 128.2, 128.1, 127.8, 113.2, 113.0, 52.4, 51.95, 50.9, 46.5. MS ESI- Found 452.34 (M - H^+), 488.24 (M + Cl^-); ESI+ Found 454.32 (M + H^+), 476.28(M + Na^+).

Compound 11C: Yield: 93%. ^{1}H NMR (400 MHz, Acetonitrile-d_3) δ 7.91 – 7.85 (m, 2H), 7.59 (dddd, *J* = 9.7, 8.2, 5.9, 2.5 Hz, 1H), 7.27 (d, *J* = 8.2 Hz, 2H), 7.21 – 7.13 (m, 2H), 7.01 – 6.94 (m, 2H), 4.54 (s, 2H), 4.47 (s, 2H). ^{19}F NMR (376 MHz, Acetonitrile-d_3) δ -116.1 (td, *J* = 9.3, 4.7 Hz), -135.3 (ddt, *J* = 21.2, 13.8, 7.2 Hz), -138.2 – -138.5 (m), -149.18 (ddd, *J* = 27.5, 19.1, 8.2 Hz), -153.4 – -154.0 (m). ^{13}C NMR (101 MHz, Acetonitrile-d_3) δ 141.3, 131.6, 130.8, 130.7, 130.4, 130.3, 129.7, 129.3, 128.4, 115.2, 115.2, 115.0, 114.9, 112.8, 112.6, 51.6, 51.6, 51.5, 49.9. MS ESI- Found 470.34 (M - H^+), 941.67 (M + Cl^-).

Compound 12C: Yield: 76%. ^{1}H NMR (400 MHz, Acetonitrile-d_3) δ 7.87 (d, *J* = 8.0 Hz, 2H), 7.67 – 7.54 (m, 1H), 7.34 – 7.20 (m, 3H), 7.05 – 6.94 (m, 2H), 6.91 (dt, *J* = 9.9, 2.2 Hz, 1H), 4.57 (s, 2H), 4.51 (s, 2H). ^{19}F NMR (376 MHz, Acetonitrile-d_3) δ -114.6 – -115.2 (m), -135.2 (ddt, *J* = 20.9, 13.8, 7.0 Hz), -137.8 – -139.0 (m), -149.2 (tt, *J* = 18.8, 8.4 Hz), -153.8 (t, *J* = 20.0 Hz). ^{13}C NMR (101 MHz, Acetonitrile-d_3) δ 141.1, 130.4, 130.3, 129.6, 129.4, 128.5, 124.4, 115.3, 115.1, 114.6, 114.4, 51.9, 51.8. MS ESI- Found 470.22 (M - H^+), 506.22 (M + Cl^-).

Compound 13C: Yield: 65%. ^{1}H NMR (400 MHz, Acetonitrile-d_3) δ 7.92 – 7.82 (m, 2H), 7.54 (dddd, *J* = 10.3, 8.2, 5.8, 2.5 Hz, 1H), 7.33 (d, *J* = 8.2 Hz, 2H), 7.30 – 7.18 (m, 2H), 7.07 (td, *J* = 7.5, 1.2 Hz, 1H), 6.94 (tt, *J* = 8.9, 1.2 Hz, 1H), 4.62 (s, 2H), 4.55 (s, 2H). ^{19}F NMR (376 MHz, Acetonitrile-d_3) δ -118.6 (dt, *J* = 12.1, 6.4 Hz), -135.4 (ddt, *J* = 20.9, 13.6, 7.2 Hz), -138.0 – -139.4 (m), -149.0 – -150.0 (m), -153.8 – -154.5 (m). ^{13}C NMR (101 MHz, Acetonitrile-d_3) δ 166.4, 141.5, 131.4, 131.3, 130.3, 130.2, 129.7, 129.3, 128.0, 124.4, 124.3, 52.1. MS ESI- Found 470.22 (M - H^+), 506.22 (M + Cl^-).

Compound 14C: Yield: 98%. ^{1}H NMR (400 MHz, DMSO-d_6) δ 8.35 (d, *J* = 4.9 Hz, 1H), 7.86 (d, *J* = 7.9 Hz, 2H), 7.83 – 7.67 (m, 2H), 7.35 (d, *J* = 7.9 Hz, 2H), 7.27 (t, *J* = 9.0 Hz, 2H), 4.67 (s, 2H), 4.61 (s, 2H). ^{19}F NMR (376 MHz, DMSO-d_6) δ -133.7 (ddt, *J* = 20.2, 13.8, 7.3 Hz), -137.3 (dt, *J* = 24.1, 11.6 Hz), -148.0 (tt, *J* = 21.2, 8.1 Hz), -153.1 (t, *J* = 21.9 Hz). ^{13}C NMR (101 MHz, DMSO-d_6) δ 167.4, 162.8, 159.0, 158.7, 155.1, 148.5, 141.2, 138.2, 130.7, 129.9, 128.7, 123.6, 123.5, 118.0, 115.1, 113.2, 113.0, 52.3, 52.1, 36.2, 31.2. MS ESI- Found 489.21 (M+Cl^-); ESI+ Found 455.22 (M+H^+) and 477.19 (M + Na^+)

TO-317-C: Yield: 88%. ^{1}H NMR (400 MHz, DMSO) δ 8.79 – 8.66 (m, 1H), 8.62 (d, *J* = 2.1 Hz, 1H), 8.27 – 8.15 (m, 1H), 7.99 – 7.85 (m, 1H), 7.88 – 7.71 (m, 3H), 7.31 (d, *J* = 8.0 Hz, 2H), 4.75 (s, 2H), 4.64 (s, 2H). ^{19}F NMR (376 MHz, DMSO) δ -133.9 (ddt, *J* = 20.7, 13.5, 7.0 Hz), -136.3 – -136.8 (m), -146.9 (tt, *J* = 22.0, 8.3 Hz), -151.8 (t, *J* = 22.1 Hz). ^{13}C NMR (101 MHz, DMSO-d_6)) δ 165.9, 150.8, 149.4, 145.3, 139.0, 136.7, 135.1, 132.4, 130.6, 129.9, 129.3, 128.5, 124.1, 112.8, 112.6, 112.4, 112.1, 51.4, 51.1, 51.0, 49.4, 49.3. MS ESI- Found 453.38 (ESI - H^+), 489.34 (M + Cl^-) and 907.54 (2M - H^+); ESI+ Found 455.17 (M + H^+).

Product D: O-(Tetrahydro-2H-pyran-2-yl) Protected Hyrodoxamate Esters:

Compound 2D: Yield: 73%. ^{1}H NMR (400 MHz, Chloroform-*d*) δ 9.41 (s, 1H), 7.73 – 7.65 (m, 2H), 7.22 (ddd, *J* = 5.0, 3.8, 2.0 Hz, 5H), 7.11 – 7.04 (m, 2H), 5.07 (t, *J* = 2.9 Hz, 1H), 4.57 (s, 2H), 4.43 (s, 2H), 4.03 (td, *J* = 10.4, 9.3, 2.8 Hz, 1H), 3.68 – 3.58 (m, 1H), 1.84 (tt, *J* = 10.1, 6.4 Hz, 3H), 1.62 – 1.52 (m, 3H). ^{19}F NMR (376 MHz, Chloroform-*d*) δ -134.6 – -135.8 (m), -146.0 (tt, *J* = 21.1, 6.5 Hz), -158.3 – -159.5 (m). ^{13}C NMR (101 MHz, Chloroform-*d*) δ 139.2, 134.0, 131.8, 128.7, 128.6, 128.5, 128.4, 127.7, 102.7, 62.6, 51.7, 51.6, 28.1, 25.0, 18.6. MS ESI- Found 569.45 (M - H^+), 605.47 (M + Cl^-), 1139.79 (2M - H^+), 1175.75 (2M + Cl^-); ESI+ Found 593.44 (M + Na^+), 1163.72 (2M + Na^+).

Compound 3D: Yield: 82%. ^{1}H NMR (400 MHz, Acetonitrile-d_3) δ 10.08 (s, 1H), 7.67 – 7.56 (m, 2H), 7.29 – 7.23 (m, 2H), 7.22 – 7.16 (m, 2H), 7.03 – 6.90 (m, 2H), 5.07 – 5.00 (m, 1H), 4.58 (s, 2H), 4.51 (s, 2H), 4.14 – 4.01 (m, 1H), 3.59 (dtd, *J* = 11.4, 3.9, 1.9 Hz, 1H), 1.84 – 1.73 (m, 3H), 1.67 – 1.50 (m, 4H). ^{19}F NMR (376 MHz, Acetonitrile-d_3) δ -115.3 – -116.3 (m), -136.8 – -137.7 (m), -148.6 (tt, *J* = 20.2, 6.9 Hz), -160.6 – -161.8 (m). ^{13}C NMR (101 MHz, Acetonitrile-d_3) δ 139.5, 131.8, 131.3, 130.8, 130.7, 128.4, 127.3, 115.2, 115.0, 101.9, 61.8, 51.9, 51.7, 27.9, 24.9, 18.3. MS: ESI- Found 587.52 (M - H^+), 623.55 (M + Cl^-), 1176.82 (2M - H^+); ESI+ Found 611.45 (M + Na^+).

Compound 4D: Yield: 77%. ^{1}H NMR (400 MHz, Chloroform-*d*) δ 9.62 (s, 1H), 7.67 – 7.59 (m, 2H), 7.44 (d, *J* = 8.1 Hz, 2H), 7.21 (s, 2H), 7.18 – 7.10 (m, 2H), 5.03 (t, *J* = 2.9 Hz, 1H), 4.53 (s, 2H), 4.49 (s, 2H), 4.00 (td, *J* = 10.1, 8.5, 2.4 Hz, 1H), 3.66 – 3.53 (m, 1H), 1.85 – 1.71 (m, 3H), 1.66 – 1.45 (m, 4H). ^{19}F NMR (376 MHz, Chloroform-*d*) δ -62.9, -132.7 – -141.4 (m), -145.4 (tt, *J* = 20.9, 6.5 Hz), -156.4 – -162.8 (m). ^{13}C NMR (101 MHz, Chloroform-*d*) δ 145.6, 142.6, 138.6, 138.5, 132.0, 128.8, 128.5, 127.7, 125.5, 125.5, 125.5, 125.4, 102.7, 62.6, 60.4, 52.0, 51.5, 28.0, 25.0, 18.6, 14.1. MS ESI- Found 637.54 (M - H^+), 673.61(M + Cl^-), 1276 (2M - H^+); ESI+ Found 661.47 (M + H^+).

Compound 5D: Yield: 69%. ^{1}H NMR (400 MHz, CD_3CN) δ 9.85 (s, 1H), 7.73 – 7.64 (m, 2H), 7.36 – 7.29 (m, 2H), 7.03 – 6.95 (m, 2H), 6.62 – 6.53 (m, 2H), 5.05 (t, *J* = 2.8 Hz, 1H), 4.63 (s, 2H), 4.39 (s, 2H), 3.61 (dtd, *J* = 11.4, 3.9, 2.0 Hz, 1H), 2.88 (s, 6H), 1.81 (h, *J* = 3.8 Hz, 3H), 1.71 – 1.53 (m, 4H). ^{19}F NMR (376 MHz, CD_3CN) δ -136.9 – -137.4 (m), -149.7 (tt, *J* = 20.4, 6.6 Hz), -161.3 – -161.9 (m). ^{13}C NMR (101 MHz, CD_3CN) δ 150.6, 140.2, 131.8, 129.9, 128.2, 127.4, 121.5, 117.3, 112.0, 101.9, 61.8, 60.0, 51.8 (d, *J* = 5.9 Hz), 39.6, 28.0, 24.9, 18.4, 13.5. MS ESI+ Found 614.43 (M + H^+), 636.43 (M + Na^+), 1249.51 (2M + Na^+); ESI- Found 612.43 (M-H^+), 648.43 (M + Cl^-).

Compound 6D: Yield: 81%. ^{1}H NMR (400 MHz, CD_3CN) δ 10.03 (s, 1H), 7.70 – 7.63 (m, 2H), 7.28 (d, *J* = 8.2 Hz, 2H), 7.06 (s, 4H), 5.05 (t, *J* = 2.9 Hz, 1H), 4.60 (s, 2H), 4.47 (s, 2H), 3.59 (ddd, *J* = 11.0, 4.2, 2.7 Hz, 1H), 2.28 (s, 3H), 1.88 – 1.72 (m, 3H), 1.70 – 1.47 (m, 3H). ^{19}F NMR (376 MHz, CD_3CN) δ -136.4 – -138.1 (m), -149.0 (tt, *J* = 20.6, 6.5 Hz), -160.8 – -162.1 (m). ^{13}C NMR (101 MHz, CD_3CN) δ 164.8, 143.3, 139.7, 139.3, 138.0, 136.8, 131.9, 131.8, 129.0, 128.7, 128.3, 127.4, 117.3, 101.9, 61.8, 60.0, 52.0, 51.8, 28.0, 25.0, 20.2, 20.1, 18.4, 13.6. MS ESI+ Found 607.36 (M + Na^+), 1191.43 (2M + Na^+); ESI- Found 583.36 (M – H^+), 619.39 (M + Cl^-), 1167.63 (2M – H^+)

Compound 7D: Yield: 65%. ^{1}H NMR (400 MHz, $CDCl_3$) δ 9.22 (s, 1H), 7.72 (s, 1H), 7.69 – 7.61 (m, 2H), 7.24 – 7.16 (m, 2H), 7.03 (d, *J* = 8.0 Hz, 2H), 6.97 (d, *J* = 8.0 Hz, 2H), 4.99 (dd, *J* = 5.5, 2.8 Hz, 1H), 3.90 (ddd, *J* = 11.1, 7.6, 3.1 Hz, 1H), 3.61 – 3.52 (m, 1H), 1.80 – 1.65 (m, 3H), 1.61 – 1.47 (m, 4H), 1.28 (s, 9H). ^{19}F NMR (376 MHz, $CDCl_3$) δ -135.0 (qd, *J* = 13.2, 7.5 Hz), -146.2 (tt, *J* = 21.1, 6.4 Hz), -158.9 (tt, *J* = 20.7, 6.3 Hz). ^{13}C NMR (101 MHz, $CDCl_3$) δ 165.7, 154.9, 151.7, 151.2, 142.6, 140.1, 139.6, 135.1, 131.8, 129.3, 128.6, 125.5, 103.1, 63.5, 60.4, 51.3, 50.9, 43.0, 43.0, 42.9, 34.7, 31.3, 29.3, 28.8, 25.1, 25.1, 21.5, 21.1, 18.7, 14.2. MS: ESI- Found 633.47 (M - H^+); ESI+ Found 657.12 (M + Na^+).

Compound 8D: Yield: 77%. ^{1}H NMR (400 MHz, CD_3CN) δ 9.95 (s, 1H), 8.44 – 8.36 (m, 2H), 7.66 – 7.55 (m, 2H), 7.28 (dd, *J* = 8.4, 2.5 Hz, 2H), 7.19 – 7.08 (m, 2H), 5.04 (t, *J* = 2.9 Hz, 1H), 4.57 (dd, *J* = 13.5, 4.5 Hz, 4H), 3.64 – 3.50 (m, 1H), 1.79 (h, *J* = 4.8, 3.6 Hz, 3H), 1.71 – 1.48 (m, 4H). ^{19}F NMR (376 MHz, CD_3CN) δ -136.7 – -137.3 (m), -148.3 (tt, *J* = 20.3, 6.8 Hz), -160.7 – -161.2 (m). ^{13}C NMR (101 MHz, CD_3CN) δ 149.6, 128.8, 127.3, 123.0, 117.3, 101.9, 61.8, 52.5, 51.6, 28.0, 24.9, 18.4. MS ESI- Found

570.64 (M - H^+), 606.67 (M + Cl^-), 1141.85 (2M - H^+) and 1177.75 (2M + Cl^-); ESI+ Found 572.53 (M + H^+), 594.47 (ESI + Na^+) and 1165.71 (2M + Na^+)

Compound 9D: Yield: 70%. ^{1}H NMR (400 MHz, Acetonitrile-d_3) δ 9.78 (s, 1H), 8.46 – 8.41 (m, 2H), 7.66 – 7.57 (m, 3H), 7.30 – 7.22 (m, 2H), 7.15 – 7.08 (m, 2H), 5.04 (d, *J* = 3.3 Hz, 1H), 4.54 (s, 2H), 4.52 (s, 2H), 3.63 – 3.57 (m, 1H), 3.49 – 3.41 (m, 1H), 1.80 (t, *J* = 2.3 Hz, 3H), 1.64 (q, *J* = 4.6 Hz, 4H). ^{19}F NMR (376 MHz, Acetonitrile-d_3) δ -135.0 (ddt, *J* = 21.1, 14.2, 6.9 Hz), -138.1 (dddd, *J* = 23.0, 13.1, 9.4, 4.1 Hz), -148.6 – -149.2 (m), -153.6 (tt, *J* = 18.2, 3.3 Hz). ^{13}C NMR (101 MHz, Acetonitrile-d_3) δ 149.8, 128.8, 127.3, 122.9, 101.9, 61.8, 60.0, 52.1, 51.2, 27.9, 24.9, 18.3, 13.5. MS ESI- Found 552.40 (M - H^+), 588.43 (M + Cl^-); ESI+ Found 554.49 (M + H^+), 576.49 (M + Na^+)

Compound 10D: Yield: 64%. ^{1}H NMR (400 MHz, Acetonitrile-d_3) δ 7.68 – 7.61 (m, 2H), 7.57 (dddd, *J* = 9.6, 8.2, 5.9, 2.5 Hz, 1H), 7.31 – 7.22 (m, 5H), 7.21 – 7.11 (m, 2H), 5.05 (t, *J* = 2.9 Hz, 1H), 4.55 (s, 2H), 4.49 (s, 2H), 3.61 (dtd, *J* = 11.4, 3.9, 1.8 Hz, 1H), 3.54 – 3.42 (m, 1H), 2.74 (s, 1H), 1.85 – 1.76 (m, 3H), 1.68 – 1.59 (m, 4H). ^{19}F NMR (376 MHz, Acetonitrile-d_3) δ -135.4 (ddt, *J* = 20.5, 13.5, 7.2 Hz), -138.0 – -138.8 (m), -149.0 – -149.7 (m), -153.9 (ddd, *J* = 21.5, 18.4, 3.3 Hz). ^{13}C NMR (101 MHz, Acetonitrile-d_3) δ 139.8, 135.3, 131.8, 128.6, 128.5, 128.4, 127.9, 127.3, 112.7, 112.5, 102.3, 101.8, 61.8, 61.79, 51.9, 51.9, 51.4, 51.4, 28.8, 28.0, 25.3, 25.2, 24.9, 19.6, 18.4, 13.5.

Compound 11D: Yield: 79%. ^{1}H NMR (400 MHz, Chloroform-*d*) δ 8.97 (s, 1H), 7.74 – 7.62 (m, 2H), 7.55 (dddd, *J* = 10.1, 8.1, 5.7, 2.5 Hz, 1H), 7.25 – 7.15 (m, 2H), 7.10 – 7.01 (m, 2H), 7.01 – 6.88 (m, 2H), 5.10 (t, *J* = 3.0 Hz, 1H), 4.49 (s, 2H), 4.39 (s, 2H), 4.03 (ddd, *J* = 11.3, 9.6, 2.9 Hz, 1H), 3.68 (dtd, *J* = 11.0, 4.0, 1.7 Hz, 1H), 1.97 – 1.79 (m, 3H), 1.65 (tdd, *J* = 12.6, 10.9, 9.9, 4.4 Hz, 4H). ^{19}F NMR (376 MHz, Chloroform-*d*) δ -113.2 (tt, *J* = 9.0, 5.0 Hz), -133.5 (ddt, *J* = 21.5, 13.8, 7.0 Hz), -134.8 – -135.4 (m), -145.7 (tt, *J* = 20.5, 8.2 Hz), -150.5 – -151.1 (m). ^{13}C NMR (101 MHz, Chloroform-*d*) δ 139.2, 131.9, 130.4, 130.3, 130.2, 128.5, 127.6, 115.8, 115.6, 112.5, 112.3, 102.8, 62.7, 50.9, 50.7, 28.1, 25.0, 18.6. MS ESI- Found 569.42, 605.39 (M + Cl^-); ESI+ Found 571.33 (M + H^+), 593.38 (M + Na^+).

Compound 12D: Yield: 87%. ^{1}H NMR (400 MHz, Acetonitrile-d_3) δ 9.77 (s, 1H), 7.66 – 7.59 (m, 2H), 7.27 (t, *J* = 7.7 Hz, 3H), 6.99 (t, *J* = 8.1 Hz, 2H), 6.92 (d, *J* = 9.9 Hz, 1H), 5.04 (s, 1H), 4.51 (dd, *J* = 17.9, 12.3 Hz, 4H), 3.03 (d, *J* = 2.7 Hz, 1H), 1.80 (d, *J* = 3.2 Hz, 3H), 1.68 – 1.58 (m, 4H). ^{19}F NMR (376 MHz, Acetonitrile-d_3) δ -114.4 – -115.2 (m), -135.2 (ddt, *J* = 21.4, 14.1, 7.5 Hz), -138.3 (dddd, *J* = 22.8, 13.1, 9.8, 4.0 Hz), -148.6 – -149.5 (m), -153.4 – -154.1 (m). ^{13}C NMR (101 MHz, Acetonitrile-d_3) δ 130.4, 130.3, 128.5, 127.3, 124.4, 124.4, 114.6, 101.9, 61.8, 60.0, 51.8, 51.6, 27.9, 24.9, 18.3, 13.5. MS ESI- Found 569.40 (M - H^+), 605.37 (M + Cl^-); 593.49 (M + Na^+).

Compound 13D: Yield: 86%. ^{1}H NMR (400 MHz, Acetonitrile-d_3) δ 9.78 (s, 1H), 5.05 (s, 1H), 4.60 (s, 2H), 4.54 (s, 2H), 3.65 – 3.58 (m, 1H), 3.02 (t, *J* = 2.6 Hz, 1H), 1.81 (d, *J* = 3.1 Hz, 3H), 1.63 (s, 4H). ^{19}F NMR (376 MHz, Acetonitrile-d_3) δ -118.2 – -119.2 (m), -135.4 (ddt, *J* = 20.6, 13.9, 7.0 Hz), -138.0 – -138.8 (m), -149.32 (ddd, *J* = 27.9, 18.6, 8.4 Hz), -153.79 – -154.37 (m). ^{13}C NMR (101 MHz, Acetonitrile-d_3) δ 140.0, 131.4, 131.3, 130.3, 130.2, 128.1, 127.3, 124.4, 124.3, 115.2, 115.0, 112.7, 112.5, 101.9, 61.8, 60.0, 52.0, 27.9, 24.9, 20.2, 18.3, 13.5. MS ESI- Found 569.40 (M - H^+), 605.37 (M + Cl^-); 593.49 (M + Na^+).

Compound 14D: Yield: 54%. ^{1}H NMR (400 MHz, Acetonitrile-d_3) δ 10.04 (s, 1H), 8.38 – 8.27 (m, 1H), 7.69 (d, *J* = 8.1 Hz, 2H), 7.60 (dtd, *J* = 17.0, 8.0, 2.2 Hz, 2H), 7.32 (d, *J* = 8.0 Hz, 2H), 7.21 – 7.08 (m, 2H), 5.05 (t, *J* = 2.8 Hz, 1H), 4.66 (s, 2H), 4.53 (s, 2H), 4.15 – 4.01 (m, 1H), 3.82 (dtd, *J* = 11.3, 8.0, 4.2 Hz, 1H), 3.65 – 3.40 (m, 2H), 1.78 (q, *J* = 8.2, 5.7 Hz, 3H), 1.62 (qt, *J* = 14.1, 10.3, 5.0 Hz, 4H). ^{19}F NMR (376 MHz, Acetonitrile-d_3) δ -134.4 (ddt, *J* = 20.6, 13.6, 7.0 Hz), -138.2 – -139.6 (m), -149.2 – -150.9 (m), -153.9 – -156.4 (m). ^{13}C NMR (101 MHz, Acetonitrile-d_3) δ 155.1, 149.1, 139.8, 136.7, 131.9, 128.4, 127.4, 125.5, 125.3, 122.9, 122.8, 117.4, 112.6, 112.6, 112.4, 112.4, 102.2, 101.9, 61.8, 61.8, 52.1, 51.6, 51.5, 28.8, 28.0, 25.2, 24.9, 19.7, 19.5, 18.4. ESI- Found 552.27 (M - H^+), 588.30 (M + Cl^-); ESI+ Found 554.40 (M + H^+), 576.39 (M + Na^+).

Compound TO-317-D: Yield: 63%. ^{1}H NMR (400 MHz, CD_3CN) δ 10.23 (s, 1H), 8.39 (dd, *J* = 4.8, 1.6 Hz, 1H), 8.30 (dd, *J* = 2.3, 0.9 Hz, 1H), 7.70 – 7.57 (m, 3H), 7.57 – 7.49 (m, 1H), 7.28 – 7.21 (m, 2H), 7.19 (ddd, *J* = 7.9, 4.8, 0.9 Hz, 1H), 5.05 (t, *J* = 2.9 Hz, 1H), 4.53 (s, 2H), 4.49 (s, 2H), 3.66 – 3.46 (m, 1H), 1.86 – 1.70 (m, 3H), 1.71 – 1.49 (m, 4H). ^{19}F NMR (376 MHz, CD_3CN) δ -134.8 – -135.3 (m), -137.7 – -138.2 (m), -148.7 (tt, *J* = 20.0, 8.1 Hz), -153.5 (ddt, *J* = 21.5, 18.5, 3.2 Hz). ^{13}C NMR (101 MHz, CD_3CN) δ

164.6, 149.5, 148.9, 139.5, 136.3, 131.9, 131.5, 128.5, 127.4, 123.4, 117.3, 112.7, 101.9, 61.8, 60.0, 52.0, 49.9, 49.8, 28.0, 24.9, 20.2, 18.4, 13.6. MS ESI- Found 552.48 ($M - H^+$), 588.52 ($M + Cl^-$), 1105.60 ($2M - H^+$) and 1141.64 ($2M + Cl^-$); ESI+ Found 554.51 ($M + H^+$),579.57 ($M + Na^+$), 1107.77 ($2M + H^+$) and 1129.74 ($2M + Na^+$).

Final Compounds: Hydroxamic Acids

The proton (^1H), fluorine (^{19}F), and carbon* (^{13}C) NMR spectra of final compounds are shown below.

Only compound peaks are shown in the following NMR reports. This is achieved by selecting the compound peak processing function using MestReNovas' advanced analysis under ^1H spectrum.

Some rotamer peaks are observed in the ^1H spectra of some compounds and are not integrated for consistency in their multiplet analysis.

Final ^{13}C spectra peaks have not been assigned integral labels, and the auto multiplet analysis function in MestReNova has been used to generate their multiplet reports.

High-Res Mass Spectras are provided as recorded from the AIMS facility at the University of Toronto and reported below to 4 decimal places.

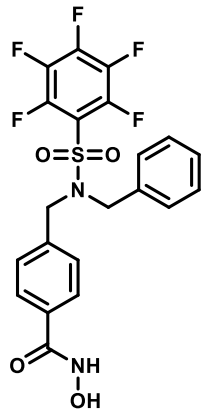
Analytical HPLC traces are provided and the purity of the final compounds as calculated by the Hewlett Packard 1100 series analytical HPLC are indicated on the spectras.

4-(((N-benzyl-2,3,4,5,6-pentafluorophenyl) sulfonamido)methyl)-N-hydroxybenzamide: Yield: 48%.

Compound 2

$C_{21}H_{15}F_5N_2O_4S$

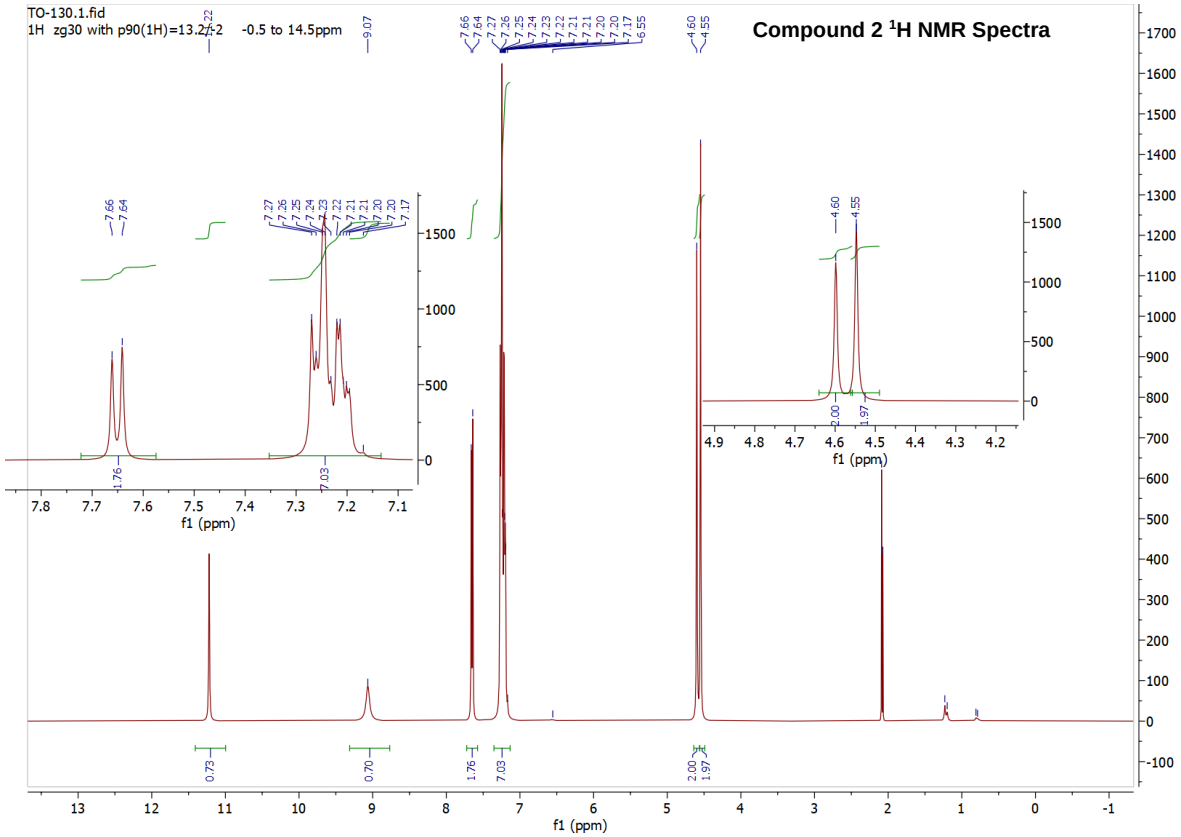
Exact Mass: 486.07

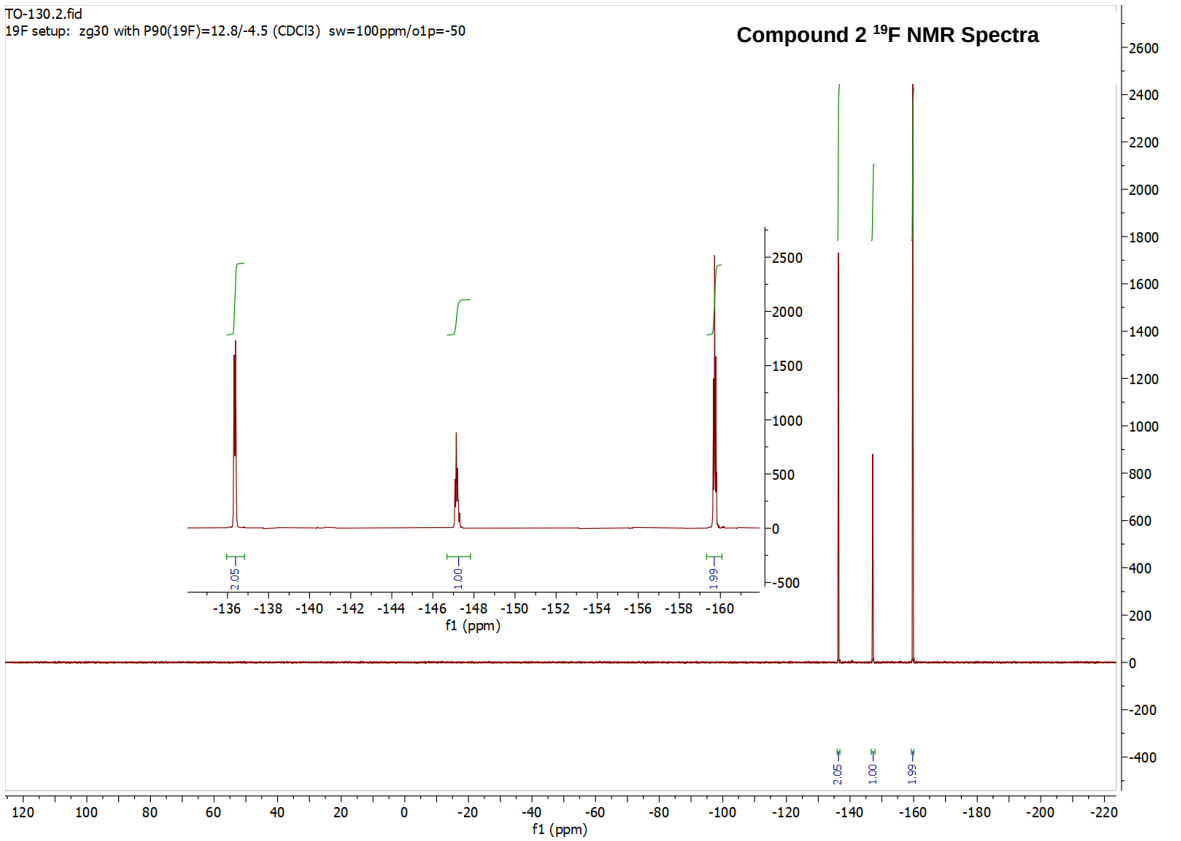


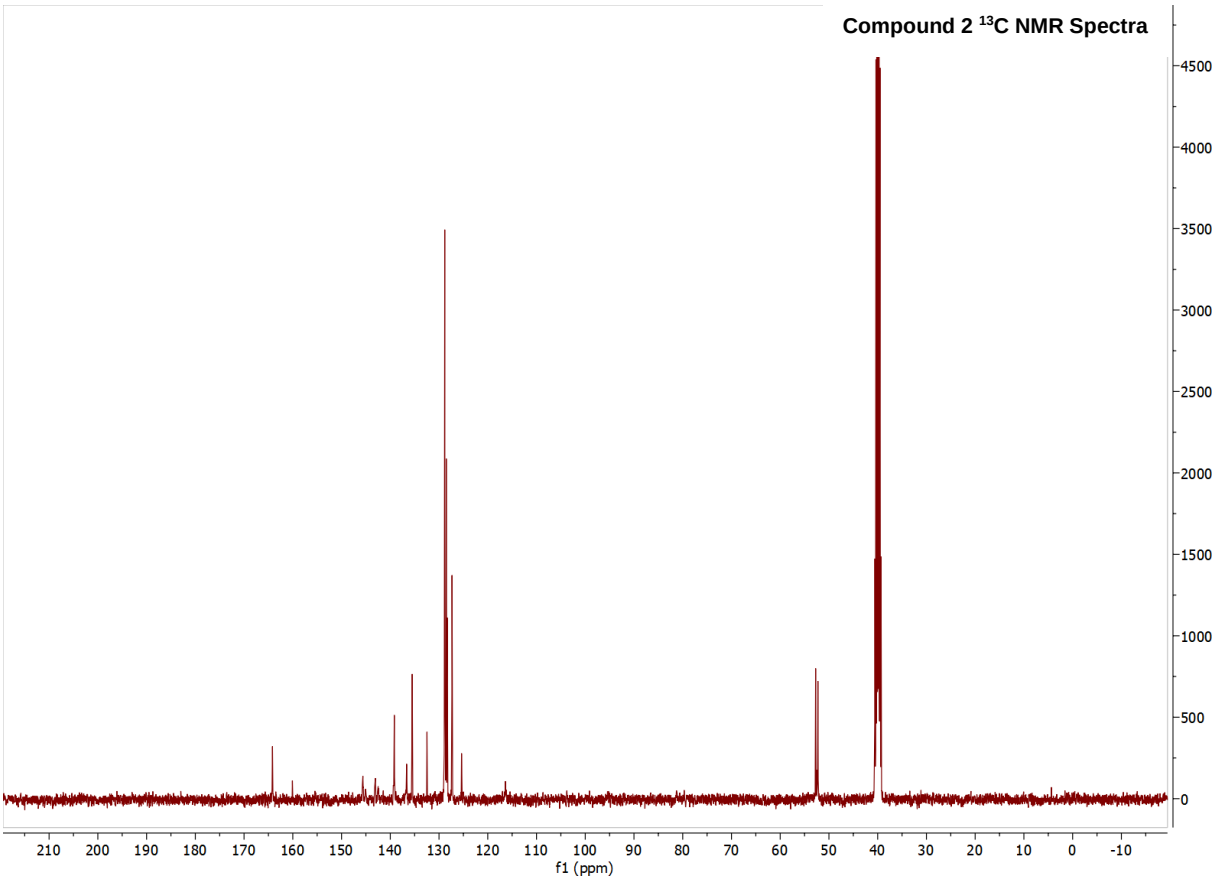
^1H NMR (400 MHz, DMSO) δ 11.22 (s, 1H), 9.07 (s, 1H), 7.65 (d, *J* = 8.0 Hz, 2H), 7.35 – 7.13 (m, 7H), 4.58 (s, 2H), 4.55 (s, 2H).

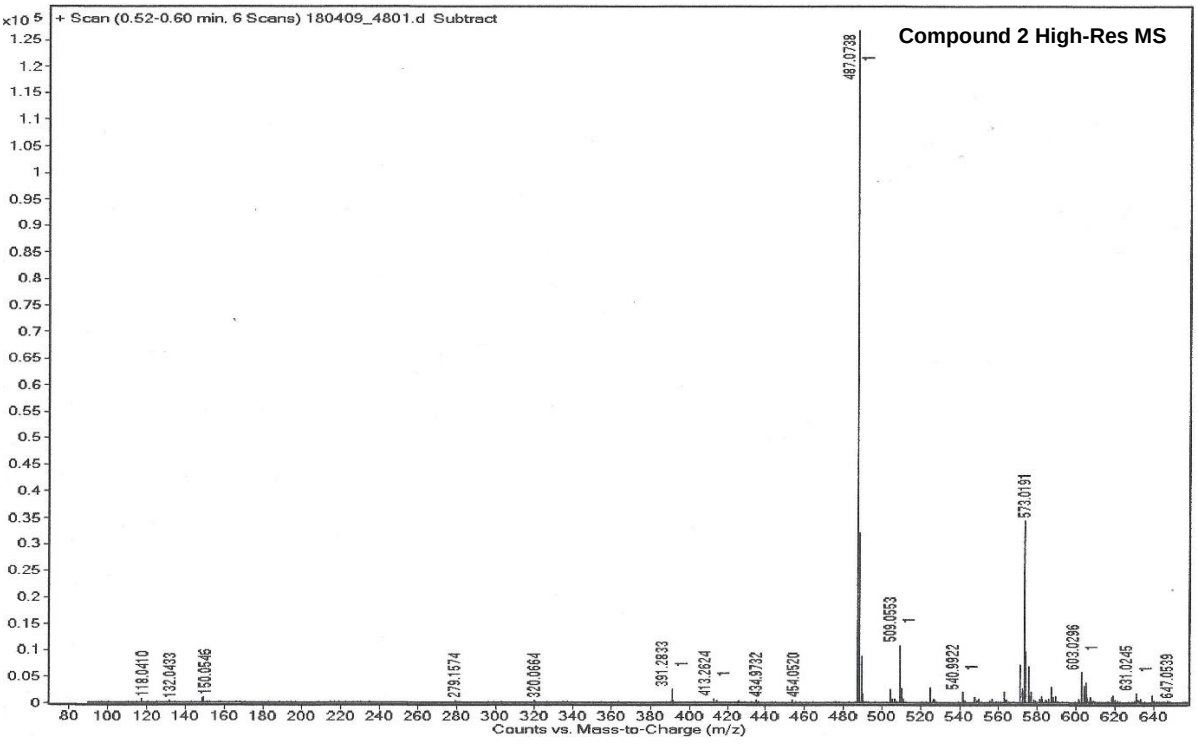
^{19}F NMR (376 MHz, DMSO) δ -136.4 (dt, *J* = 22.3, 4.9 Hz), -146.7 – -147.8 (m), -159.3 – -160.1 (m).

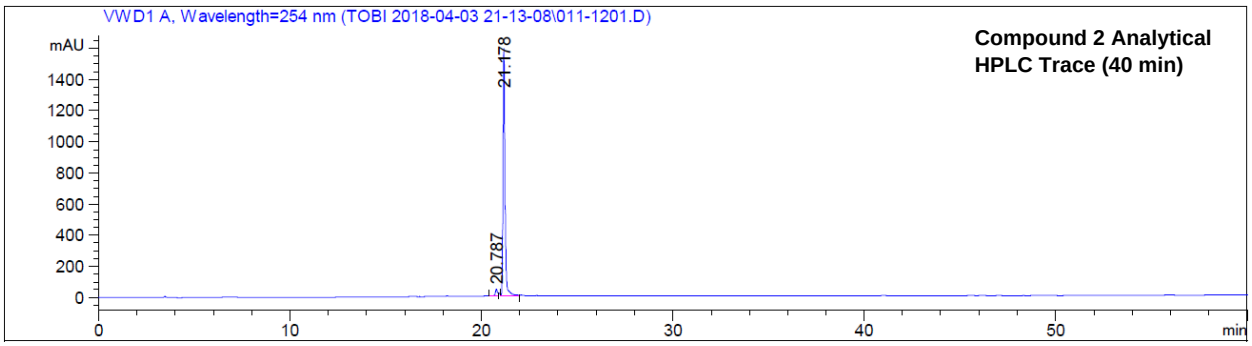
^{13}C NMR (101 MHz, DMSO) δ 164.2, 160.1, 145.6, 139.2, 136.6, 135.6, 135.5, 132.5, 129.0, 128.9, 128.8, 128.8, 128.7, 128.5, 128.3, 128.3, 127.3, 125.4, 52.7, 52.5, 52.3.


TO-130.1.fid
1H zg30 with p90(1H)=13.2/-2 -0.5 to 14.5ppm
Compound 2 ^{1}H NMR Spectra
f1 (ppm)

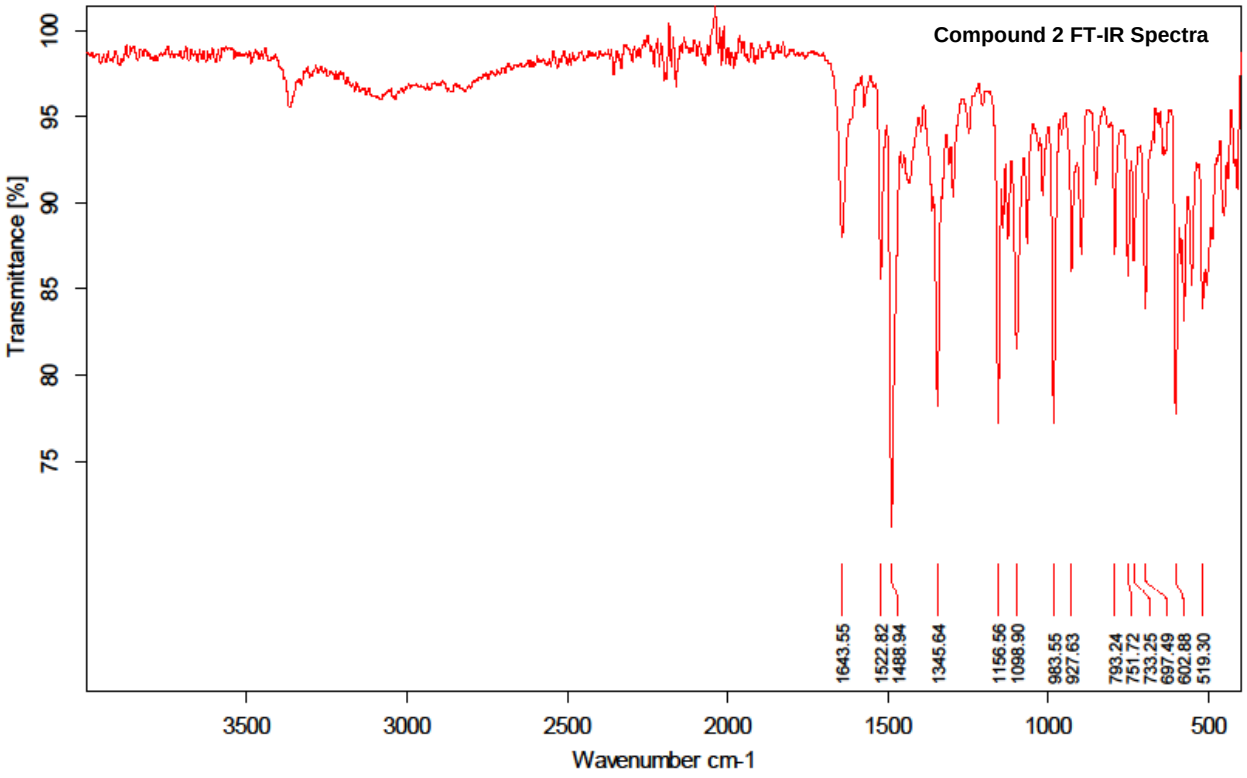

TO-130.2.fid
19F setup: zg30 with P90(19F)=12.8/-4.5 (CDCl3) sw=100ppm/o1p=-50
Compound 2 ^{19}F NMR Spectra
f1 (ppm)


Compound 2 ^{13}C NMR Spectra
f1 (ppm)


Compound 2 High-Res MS
+ Scan (0.52-0.60 min, 6 Scans) 180409_4801.d Subtract
Counts vs. Mass-to-Charge (m/z)



Compound 2 Analytical HPLC Trace (40 min)



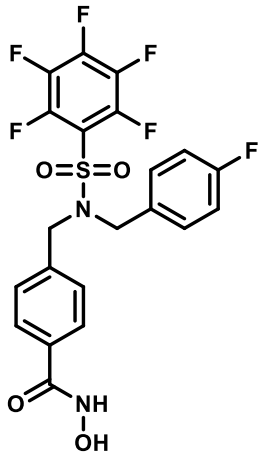
Compound 2 FT-IR Spectra

N-hydroxy-4-(((2,3,4,5,6-pentafluoro-N-(4-fluorobenzyl)phenyl)sulfonamido)methyl)benzamide
Yield: 53%

Compound 3

$C_{21}H_{14}F_6N_2O_4S$

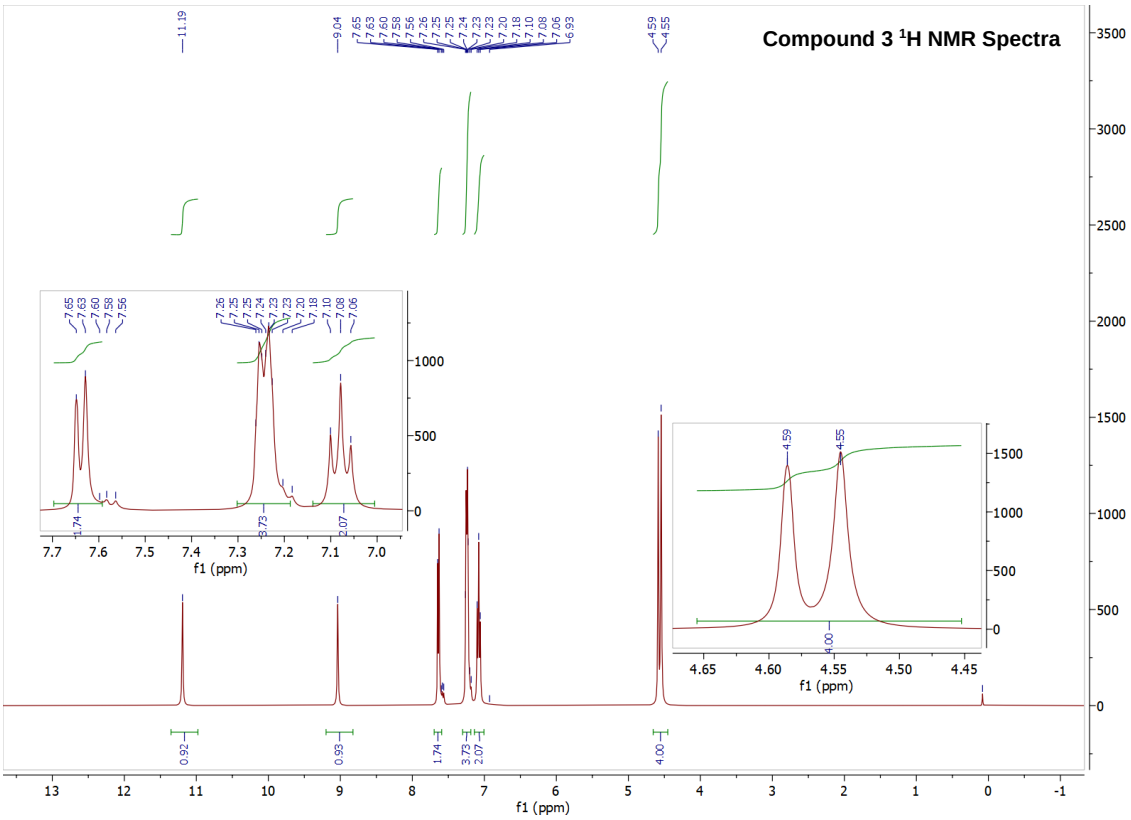
Exact Mass: 504.06

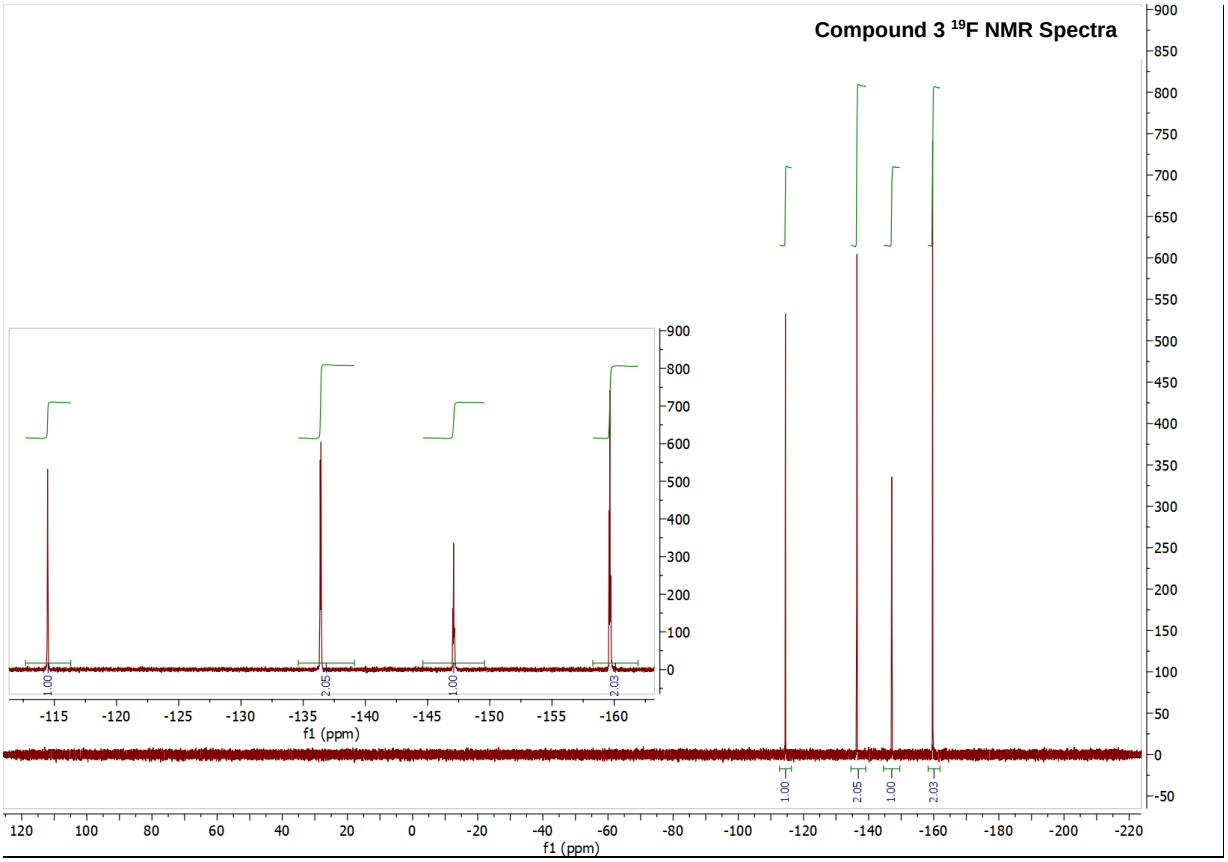


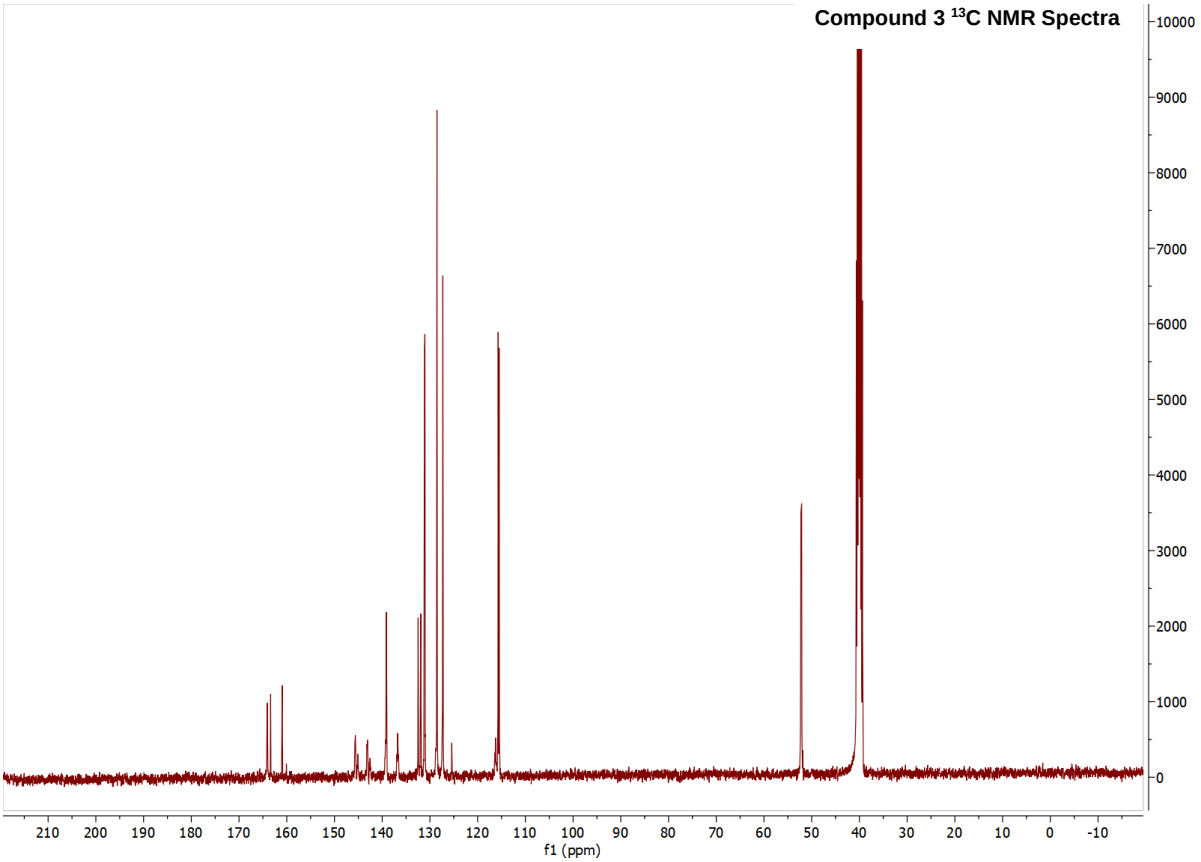
^{1}H NMR (400 MHz, DMSO) δ 11.19 (s, 1H), 9.04 (s, 1H), 7.64 (d, *J* = 7.8 Hz, 2H), 7.24 (dt, *J* = 9.0, 2.8 Hz, 4H), 7.08 (t, *J* = 8.8 Hz, 2H), 4.57 (d, *J* = 16.3 Hz, 4H).

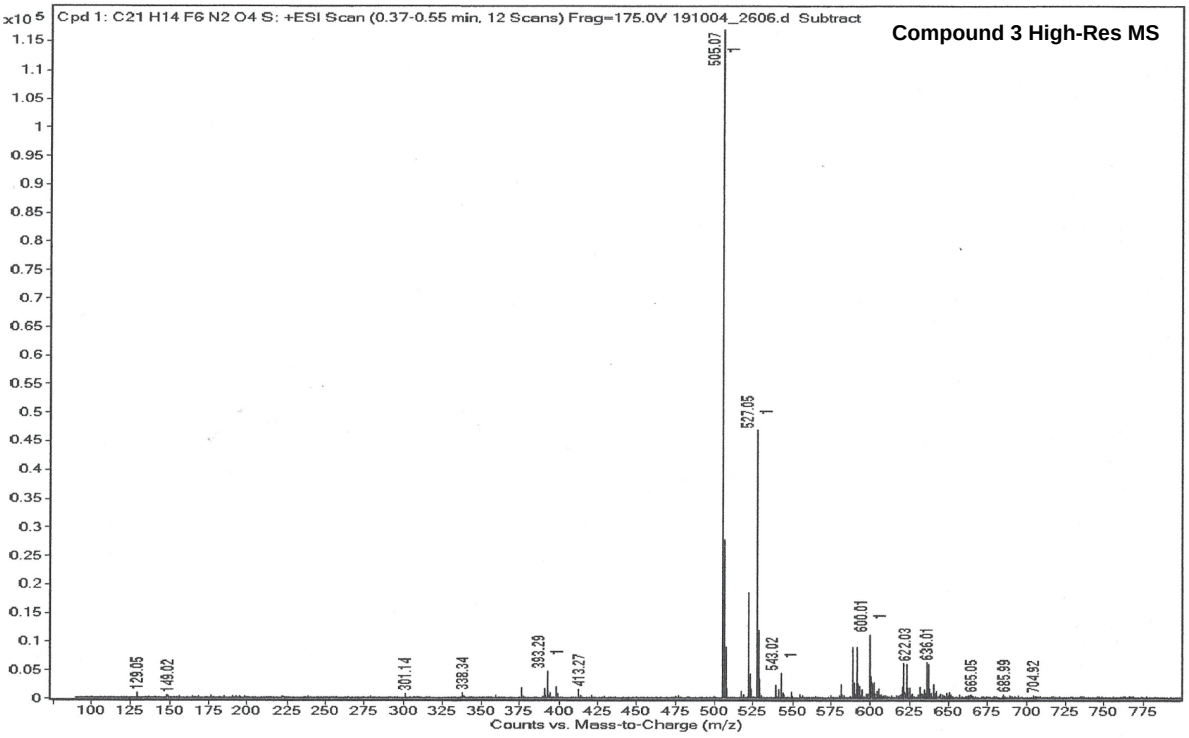
^{19}F NMR (376 MHz, DMSO) δ -114.5 (tq, *J* = 12.8, 7.0, 6.1 Hz), -136.4 (dt, *J* = 23.3, 5.1 Hz), -147.1 (tt, *J* = 22.7, 6.5 Hz), -159.7 (tt, *J* = 22.6, 5.8 Hz).

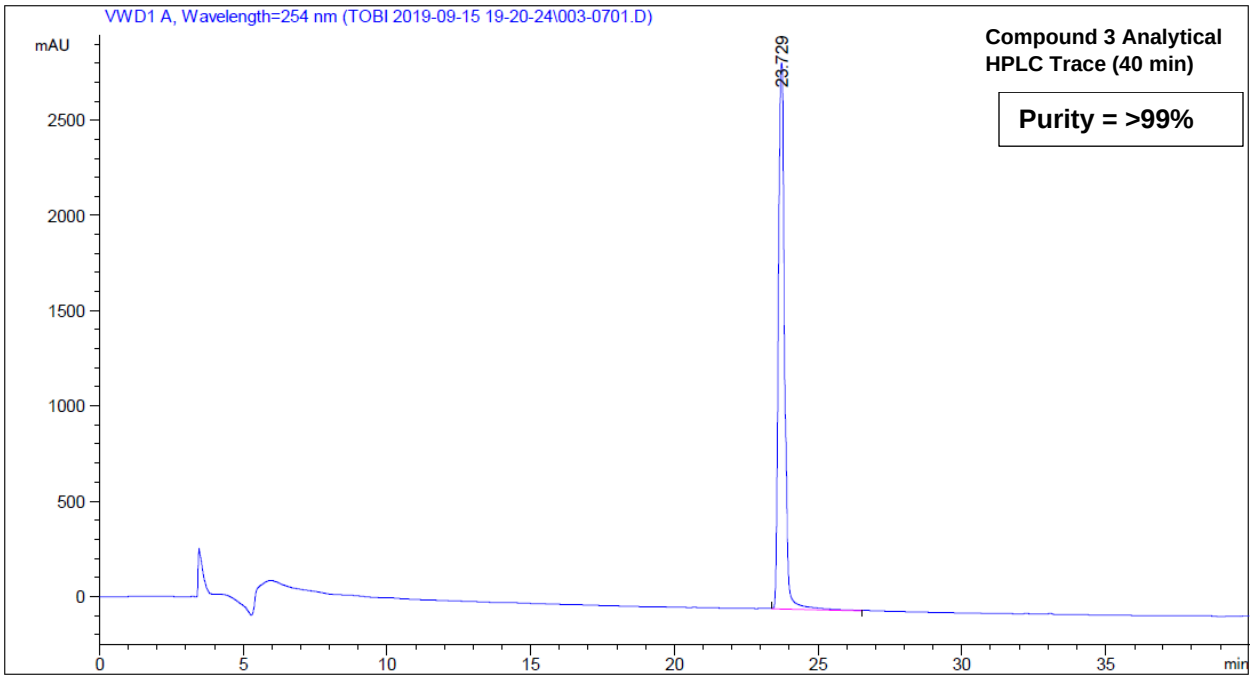
^{13}C NMR (101 MHz, DMSO) δ 164.3, 145.8, 139.3, 136.9, 132.1, 131.2, 128.7, 128.6, 127.5, 127.4, 116.4, 115.9, 115.8, 115.7, 115.6, 52.4, 52.3, 40.8, 40.6, 40.4.

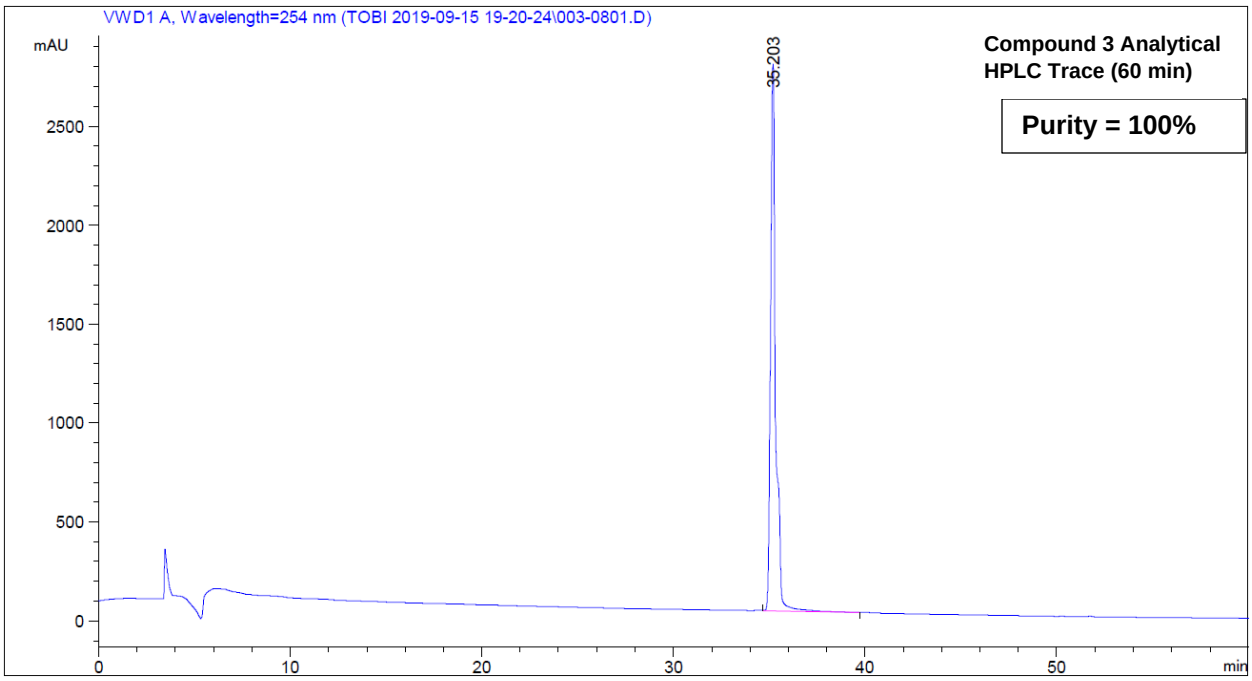


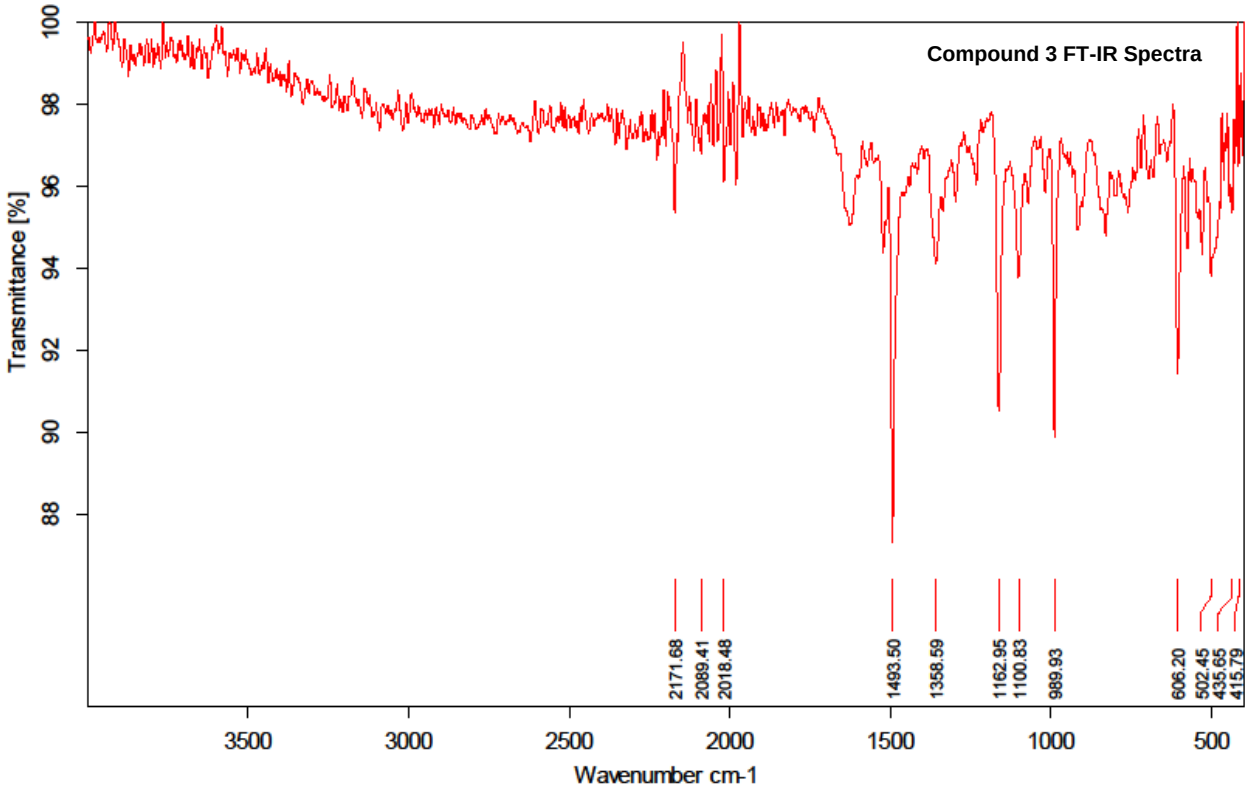

Compound 3 19F NMR Spectra
f1 (ppm)
1.00
2.05
1.00
2.03


Compound 3 13C NMR Spectra
f1 (ppm)


Cpd 1: C21 H14 F6 N2 O4 S: +ESI Scan (0.37-0.55 min, 12 Scans) Frag=175.0V 191004_2606.d Subtract
Compound 3 High-Res MS
x10 5
505.07
527.05
600.01
622.03
636.01
393.29
543.02
129.05
148.02
301.14
338.34
413.27
665.05
685.99
704.92
Counts vs. Mass-to-Charge (m/z)


VWD1 A, Wavelength=254 nm (TOBI 2019-09-15 19-20-24\003-0701.D)
mAU
23.729
Compound 3 Analytical HPLC Trace (40 min)
Purity = >99%
min


VWD1 A, Wavelength=254 nm (TOBI 2019-09-15 19-20-24\003-0801.D)
mAU
2500
2000
1500
1000
500
0
35.203
0
10
20
30
40
50
min
Compound 3 Analytical HPLC Trace (60 min)
Purity = 100%

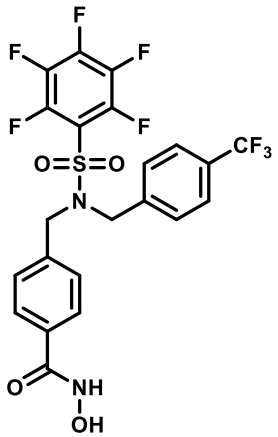

Compound 3 FT-IR Spectra
Transmittance [%]
100
98
96
94
92
90
88
2171.68
2089.41
2018.48
1493.50
1358.59
1162.95
1100.83
989.93
606.20
502.45
435.65
415.79
3500
3000
2500
2000
1500
1000
500
Wavenumber cm-1

N-hydroxy-4-(((2,3,4,5,6-pentafluoro-N-(4-(trifluoromethyl)benzyl)phenyl)sulfonamido)methyl)benzamide Yield: 54%

Compound 4

$C_{22}H_{14}F_8N_2O_4S$

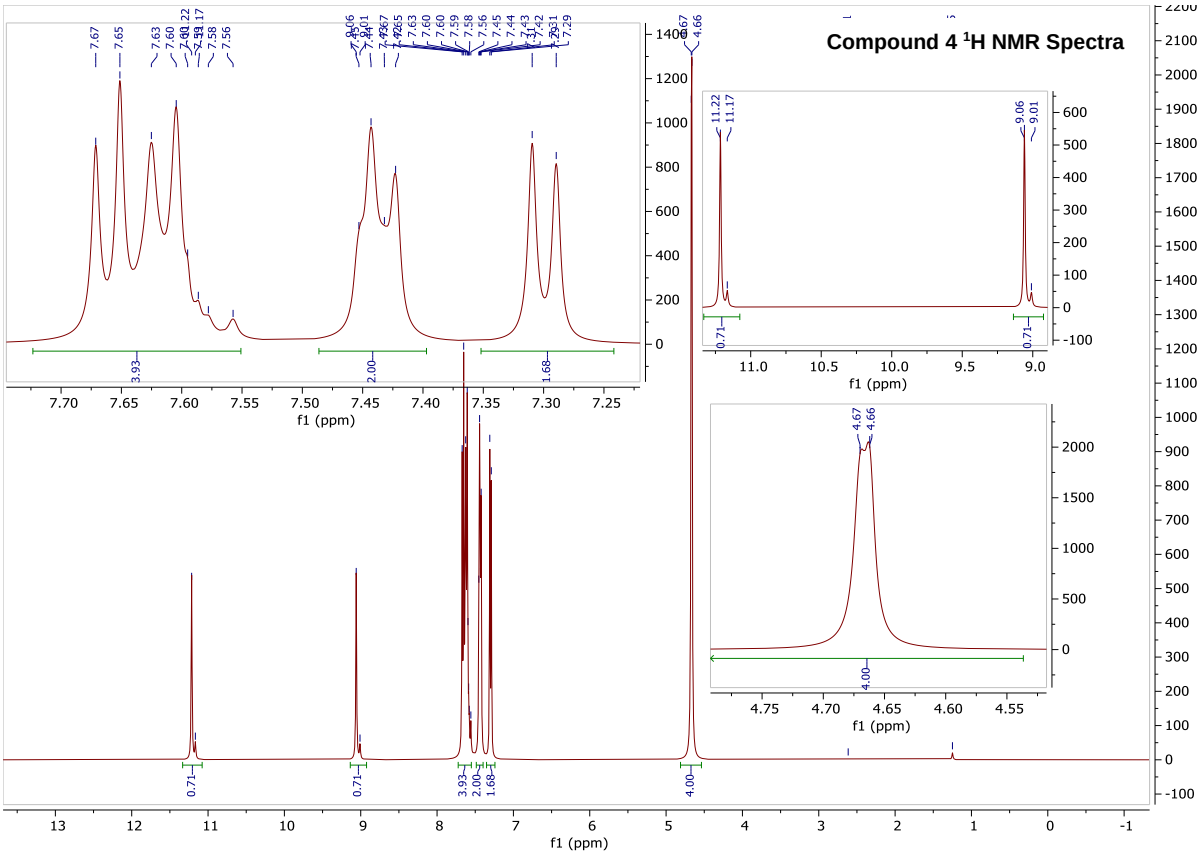
Mol. Weight: 554.41 $gmol^{-1}$

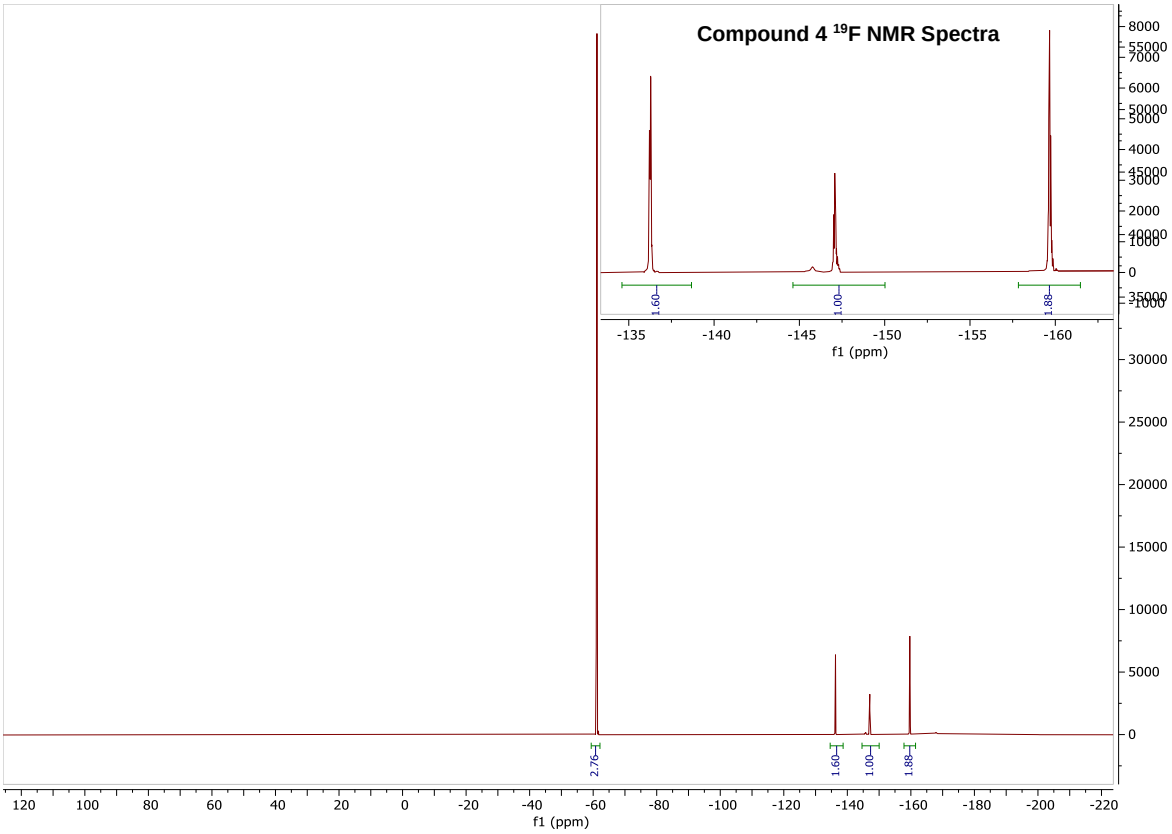


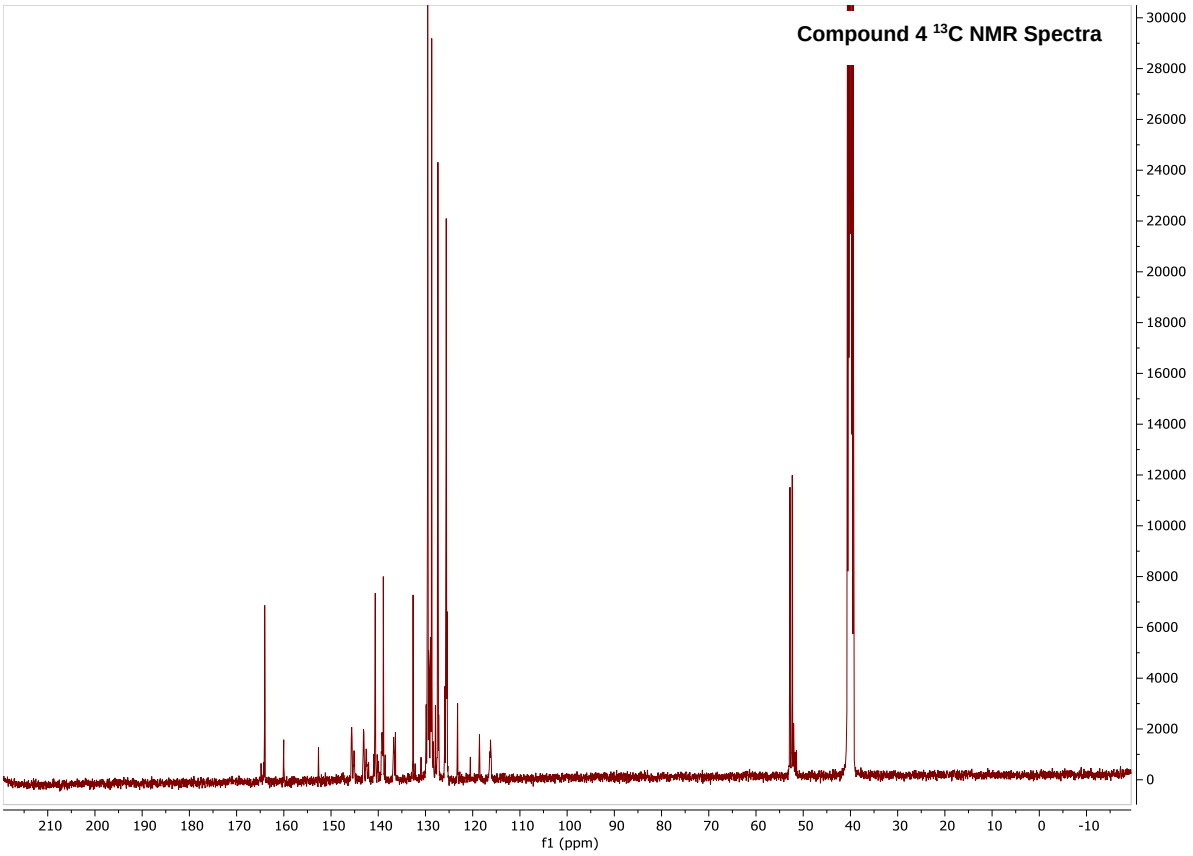
^{1}H NMR (400 MHz, DMSO) δ 11.22 (s, 1H), 9.06 (s, 1H), 7.72 – 7.55 (m, 4H), 7.44 (dd, *J* = 8.3, 3.9 Hz, 2H), 7.30 (d, *J* = 8.0 Hz, 2H), 4.67 (d, *J* = 3.2 Hz, 4H).

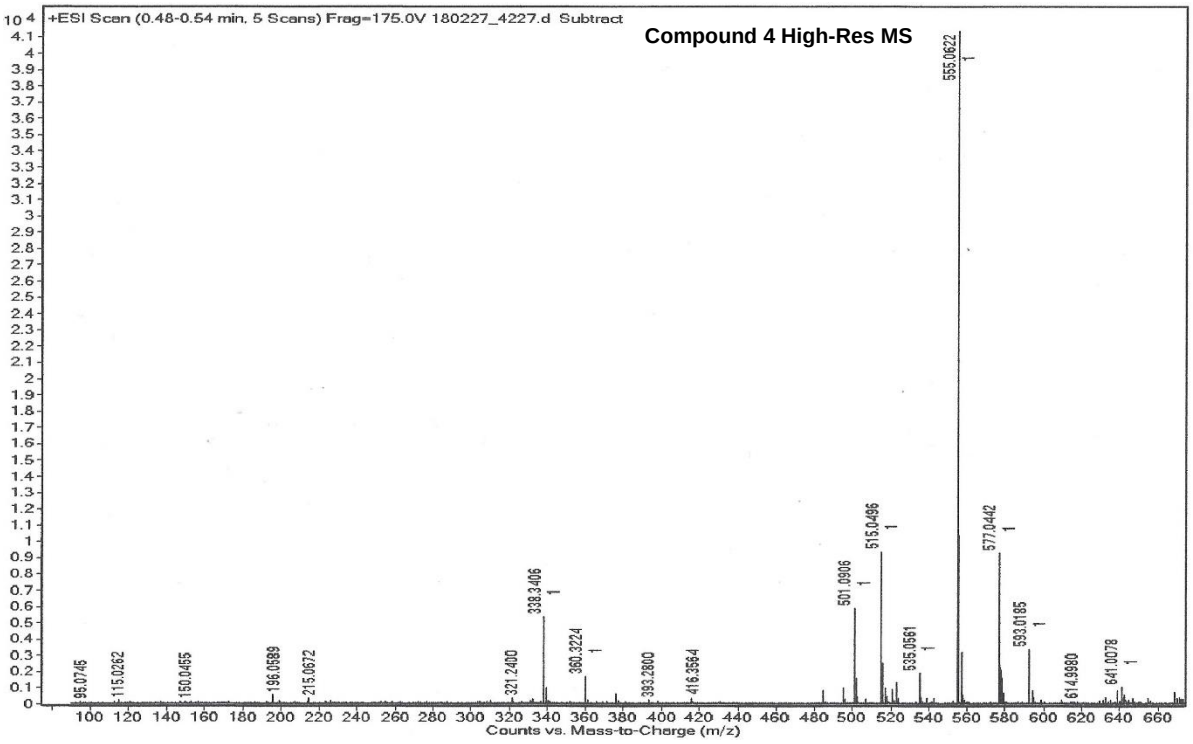
^{19}F NMR (376 MHz, DMSO) δ -59.4 – -62.1 (m), -136.3 (dq, *J* = 30.5, 12.5, 9.6 Hz), -144.6 – -150.0 (m), -157.8 – -161.5 (m).

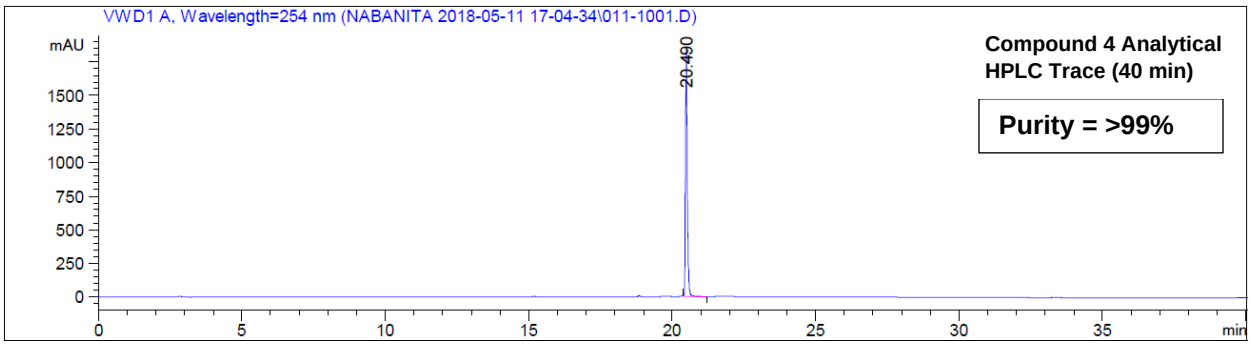
^{13}C NMR (101 MHz, DMSO) δ 164.3, 160.0, 152.7, 145.6, 145.1, 142.6, 140.8, 139.2, 138.9, 136.7, 132.6, 129.9, 129.0, 127.9, 125.9, 123.2, 118.6, 52.8

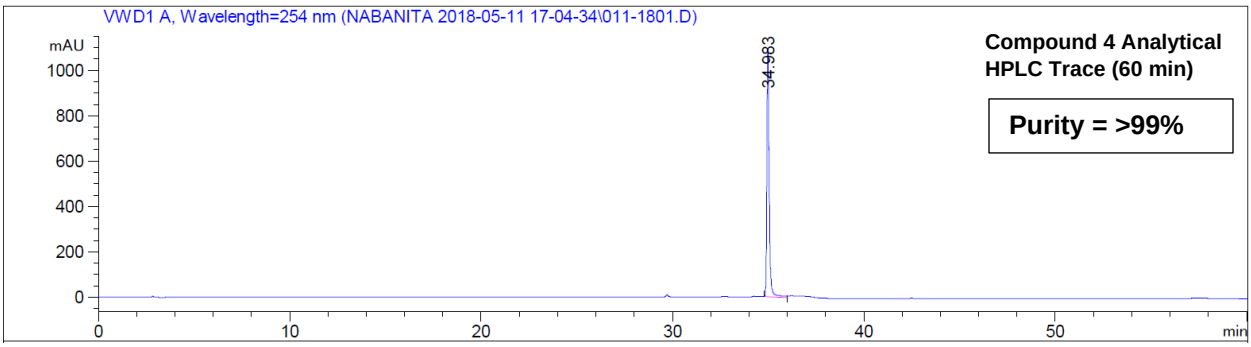

Compound 4 ^{1}H NMR Spectra
f1 (ppm)

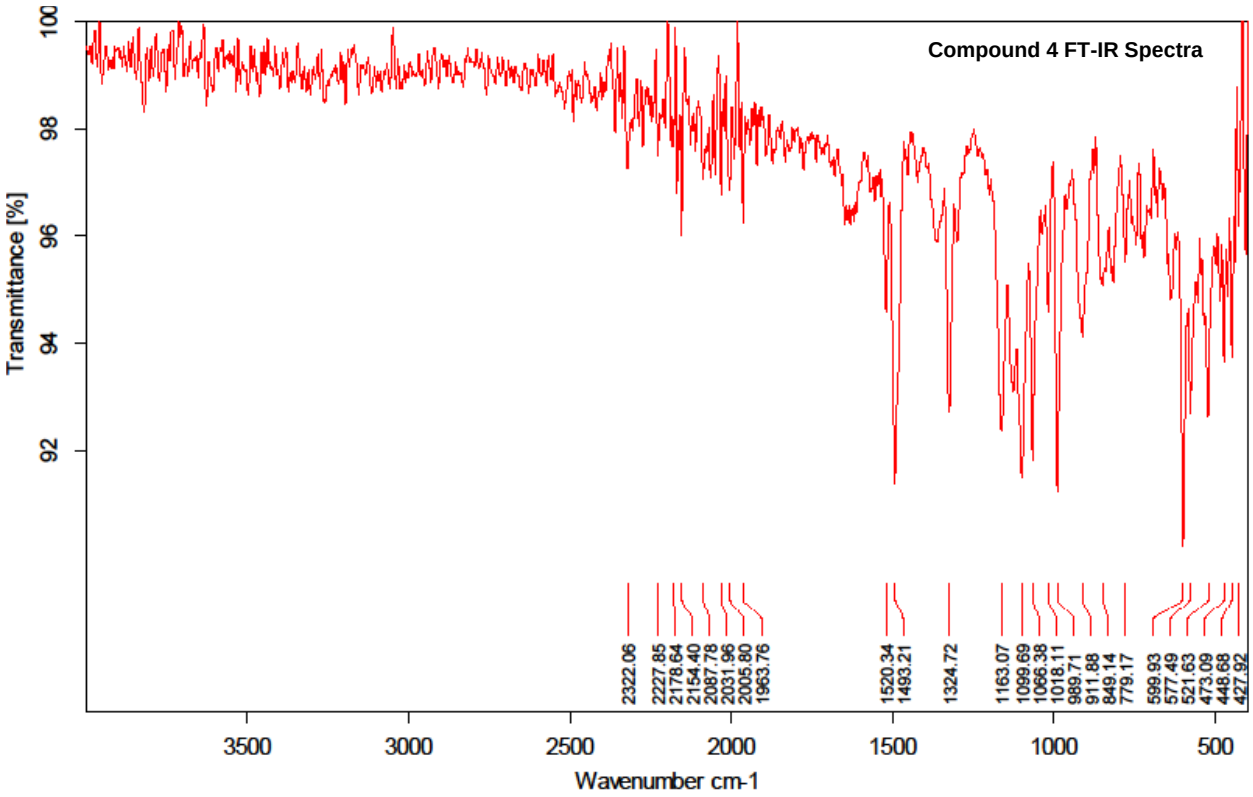

Compound 4 ^{19}F NMR Spectra
f1 (ppm)


Compound 4 13C NMR Spectra
f1 (ppm)


Compound 4 High-Res MS
+ESI Scan (0.48-0.54 min, 5 Scans) Frag=175.0V 180227_4227.d Subtract
Counts vs. Mass-to-Charge (m/z)


VWD1 A, Wavelength=254 nm (NABANITA 2018-05-11 17-04-34\011-1001.D)
mAU
1500
1250
1000
750
500
250
0
20.490
0
5
10
15
20
25
30
35
min
Compound 4 Analytical
HPLC Trace (40 min)
Purity = >99%


VWD1 A, Wavelength=254 nm (NABANITA 2018-05-11 17-04-34\011-1801.D)
mAU
1000
800
600
400
200
0
34.983
0
10
20
30
40
50
min
Compound 4 Analytical
HPLC Trace (60 min)
Purity = >99%

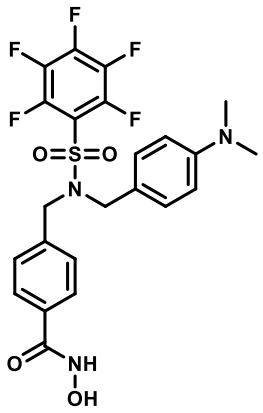

Compound 4 FT-IR Spectra
Transmittance [%]
100
98
96
94
92
2322.06
2227.85
2178.64
2154.40
2087.78
2031.96
2005.80
1963.76
1520.34
1493.21
1324.72
1163.07
1099.69
1066.38
1018.11
989.71
911.88
849.14
779.17
599.93
577.49
521.63
473.09
448.68
427.92
3500
3000
2500
2000
1500
1000
500
Wavenumber cm-1

4-(((N-(4-(dimethylamino)benzyl)-2,3,4,5,6-pentafluorophenyl)sulfonamido)methyl)-N-hydroxybenzamide. Yield: 38%

Compound 5

$C_{23}H_{20}F_5N_3O_4S$

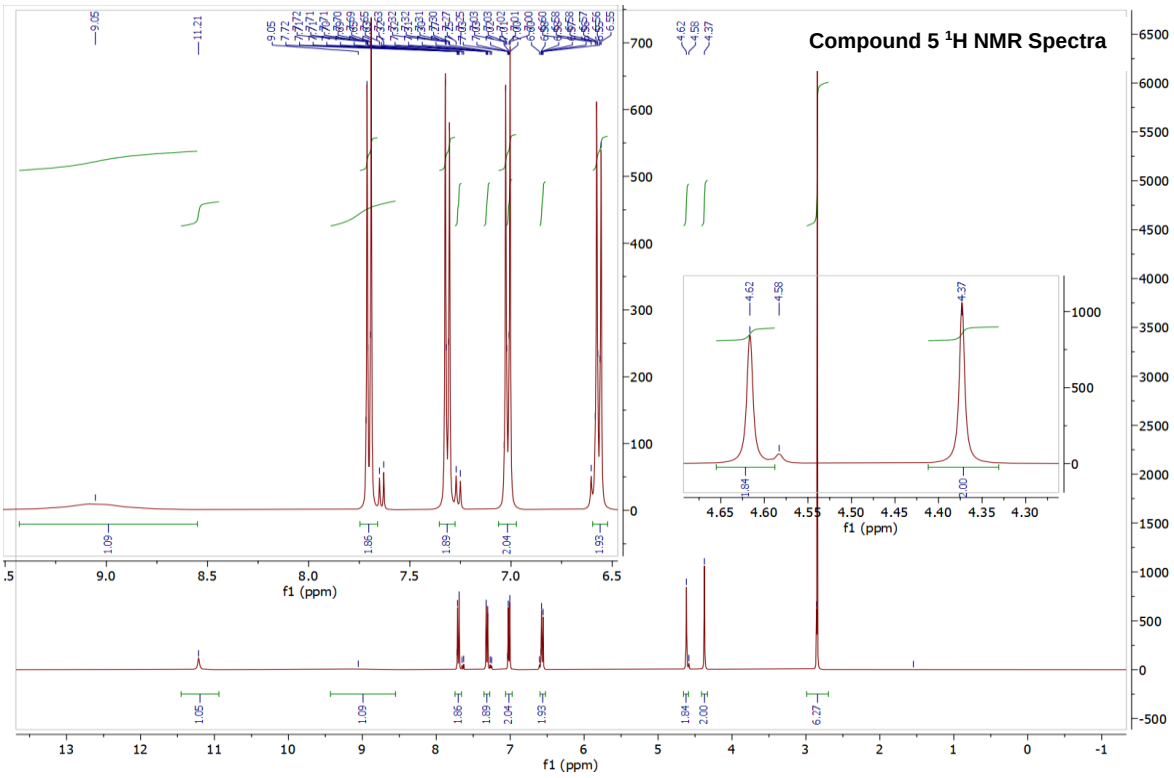
Mol. Weight: 529.49 $gmol^{-1}$

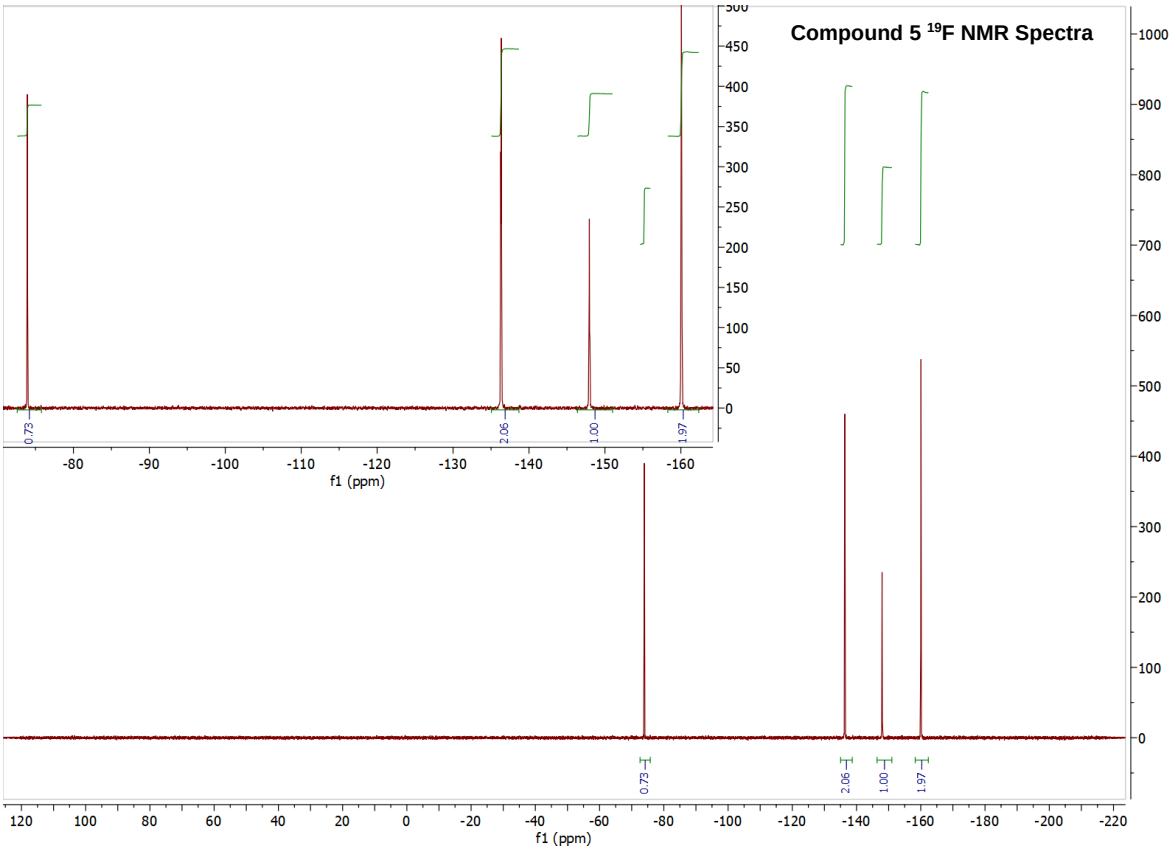


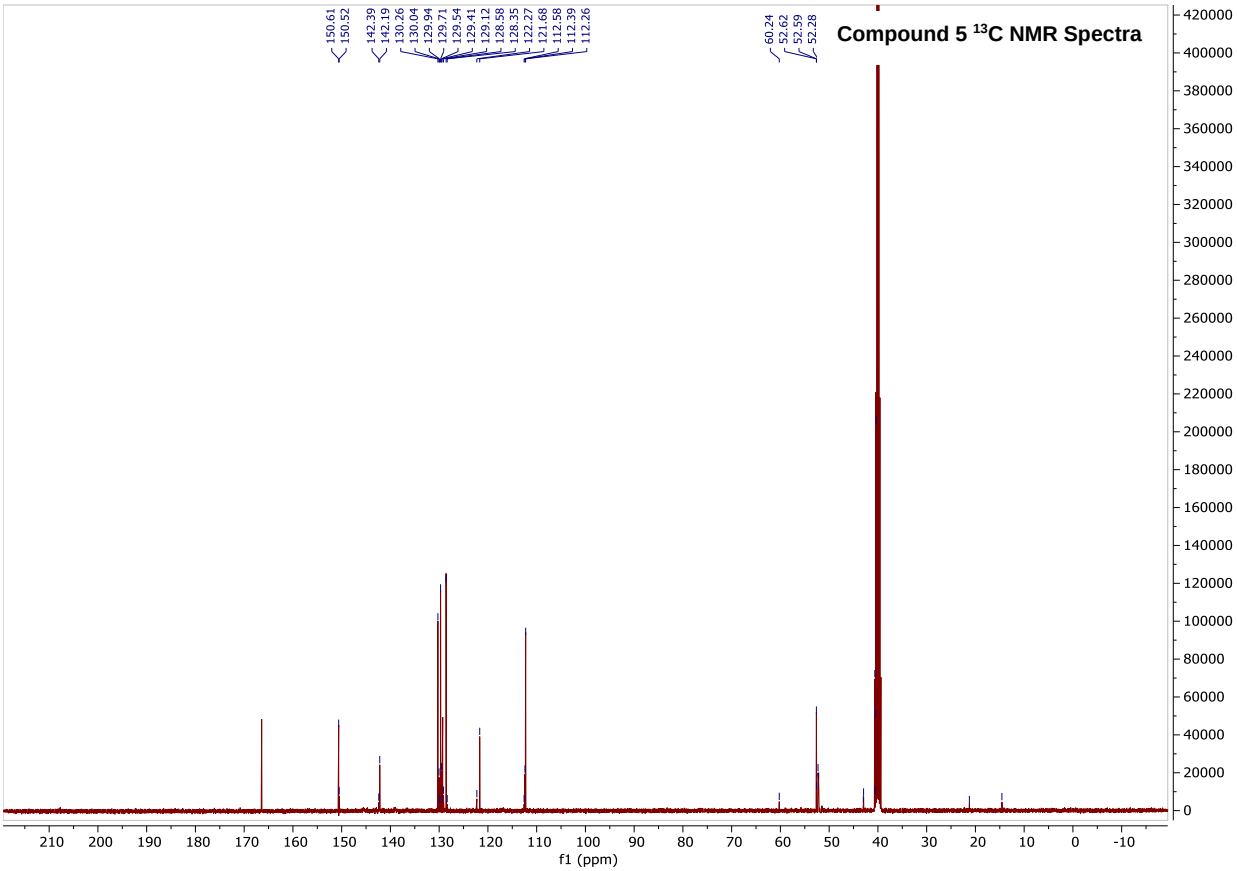
^{1}H NMR (400 MHz, DMSO) δ 11.21 (s, 1H), 9.05 (s, 1H), 7.75 – 7.66 (m, 2H), 7.35 – 7.28 (m, 2H), 7.06 – 6.97 (m, 2H), 6.60 – 6.52 (m, 2H), 4.62 (s, 2H), 4.37 (s, 2H), 2.85 (s, 6H).

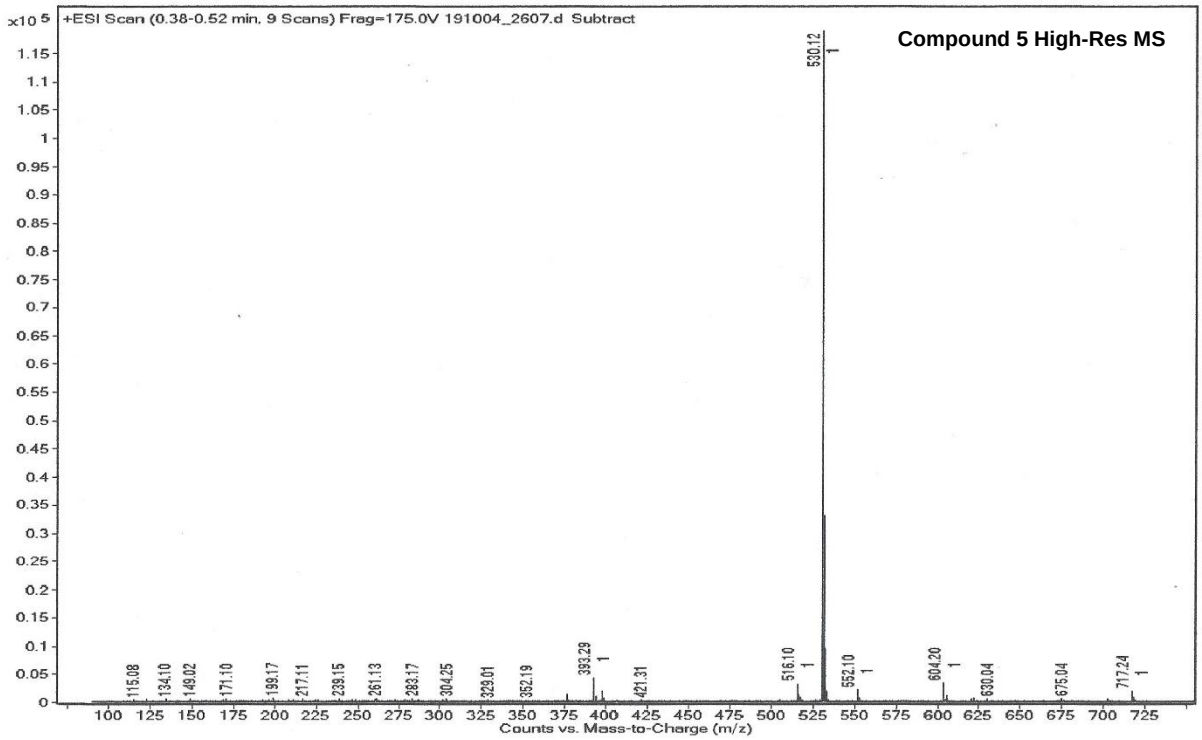
^{19}F NMR (376 MHz, DMSO) δ -73.9, -136.3 (d, *J* = 24.0 Hz), -148.0 (t, *J* = 23.2 Hz), -160.1 (t, *J* = 21.8 H)

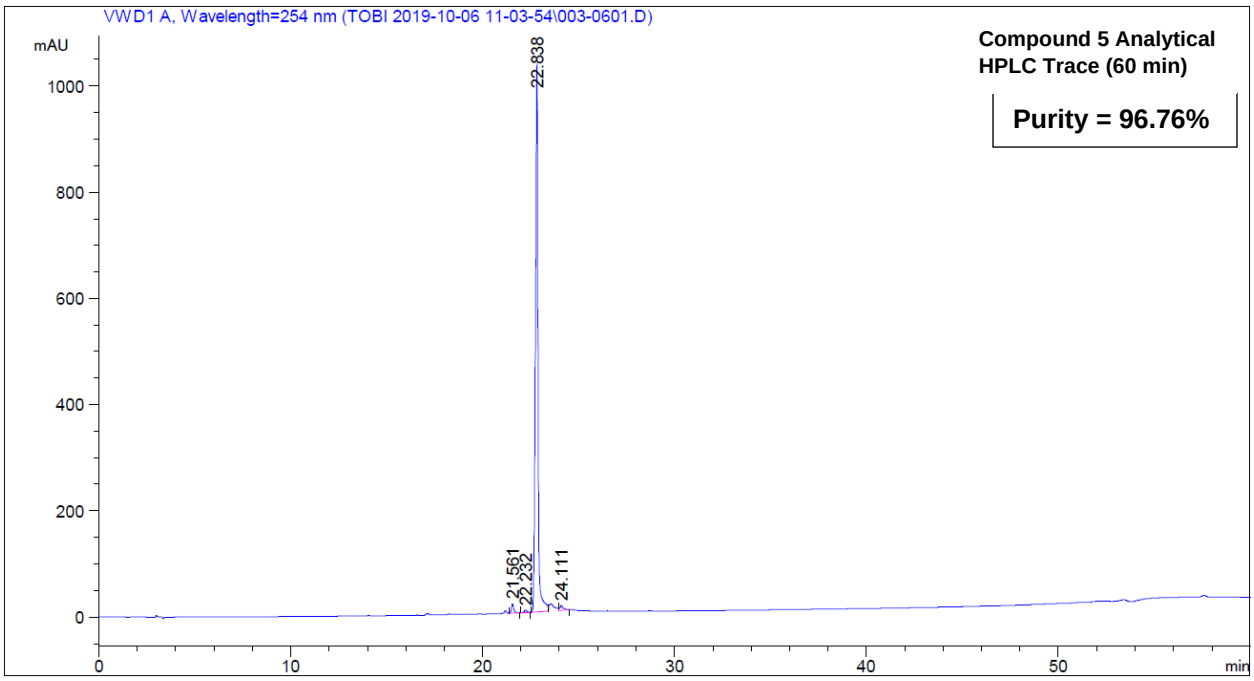
^{13}C NMR (101 MHz, DMSO) δ 166.4, 150.6, 142.6, 142.4, 142.2, 136.6, 130.3, 130.0, 129.9, 129.7, 129.5, 129.4, 129.3, 129.1, 128.6, 122.3, 121.7, 116.9, 112.4, 112.3, 60.2, 52.6, 52.6, 52.3, 52.1, 51.5, 42.9, 42.9, 14.6.




Compound 5 ^{19}F NMR Spectra
f1 (ppm)
0.73
2.06
1.00
1.97


Compound 5 ^{13}C NMR Spectra
150.61
150.52
142.39
142.19
130.26
130.04
129.94
129.71
129.54
129.41
129.12
128.58
128.35
122.27
121.68
112.58
112.39
112.26
60.24
52.62
52.59
52.28
f1 (ppm)


+ESI Scan (0.38-0.52 min, 9 Scans) Frag=175.0V 191004_2607.d Subtract
x10 5
Compound 5 High-Res MS
530.12
Counts vs. Mass-to-Charge (m/z)

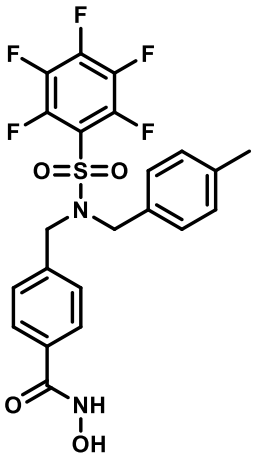

VWD1 A, Wavelength=254 nm (TOBI 2019-10-06 11-03-54\003-0601.D)
mAU
Compound 5 Analytical HPLC Trace (60 min)
Purity = 96.76%
22.838
21.561
22.232
24.111
min

N-hydroxy-4-(((2,3,4,5,6-pentafluoro-N-(4-methylbenzyl)phenyl)sulfonamido)methyl)benzamide.
Yield: 61%

Compound 6

$C_{22}H_{17}F_5N_2O_4S$

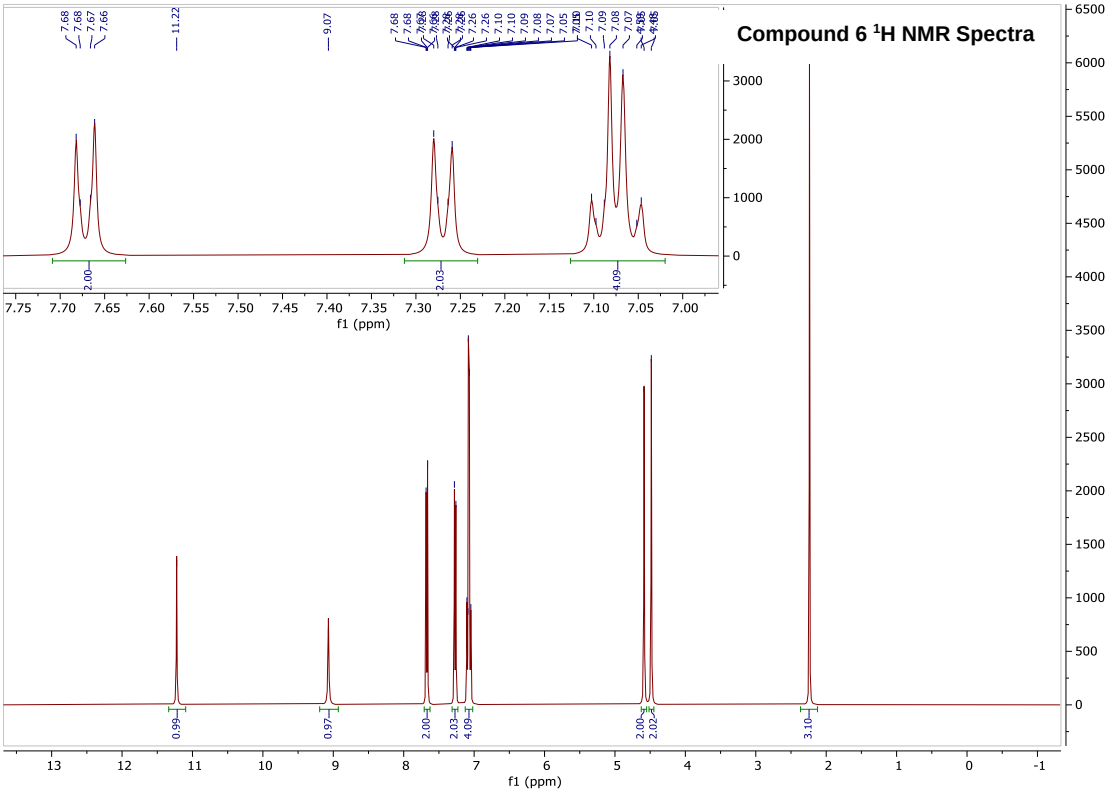
Mol. Weight: 500.45 $gmol^{-1}$

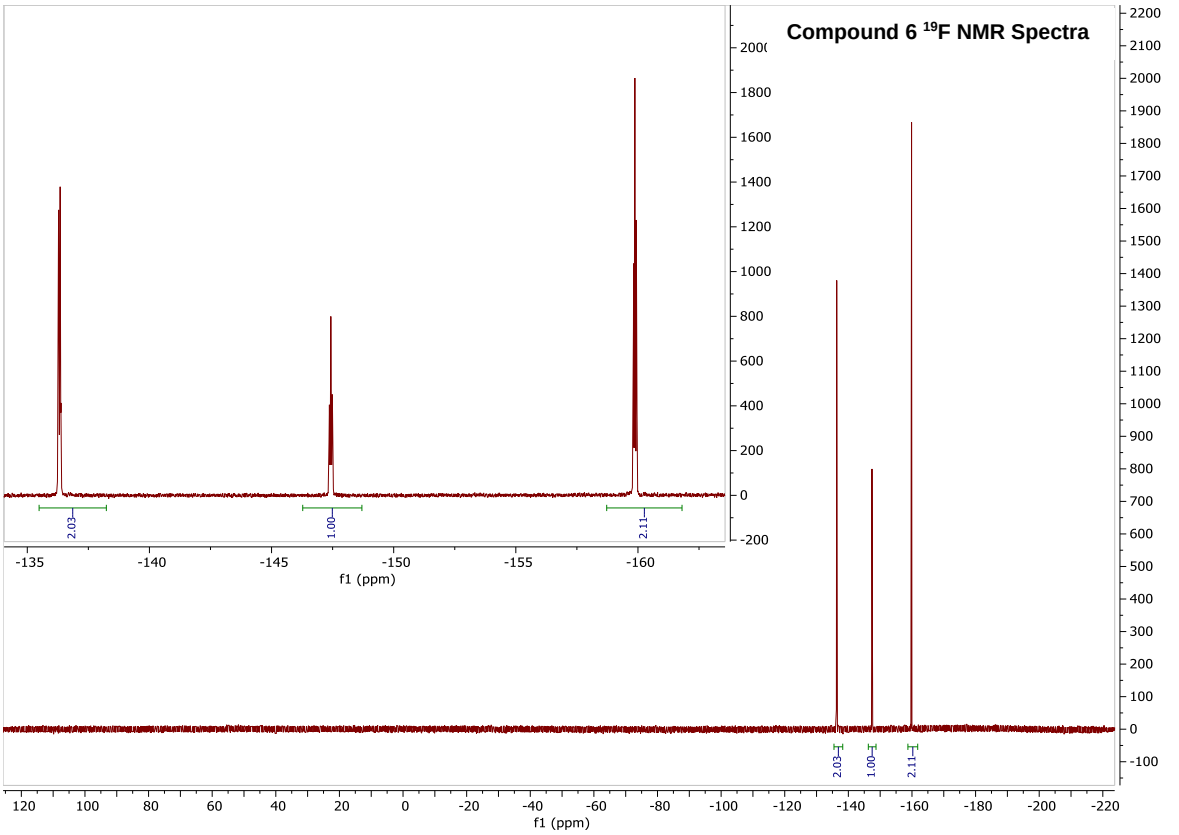


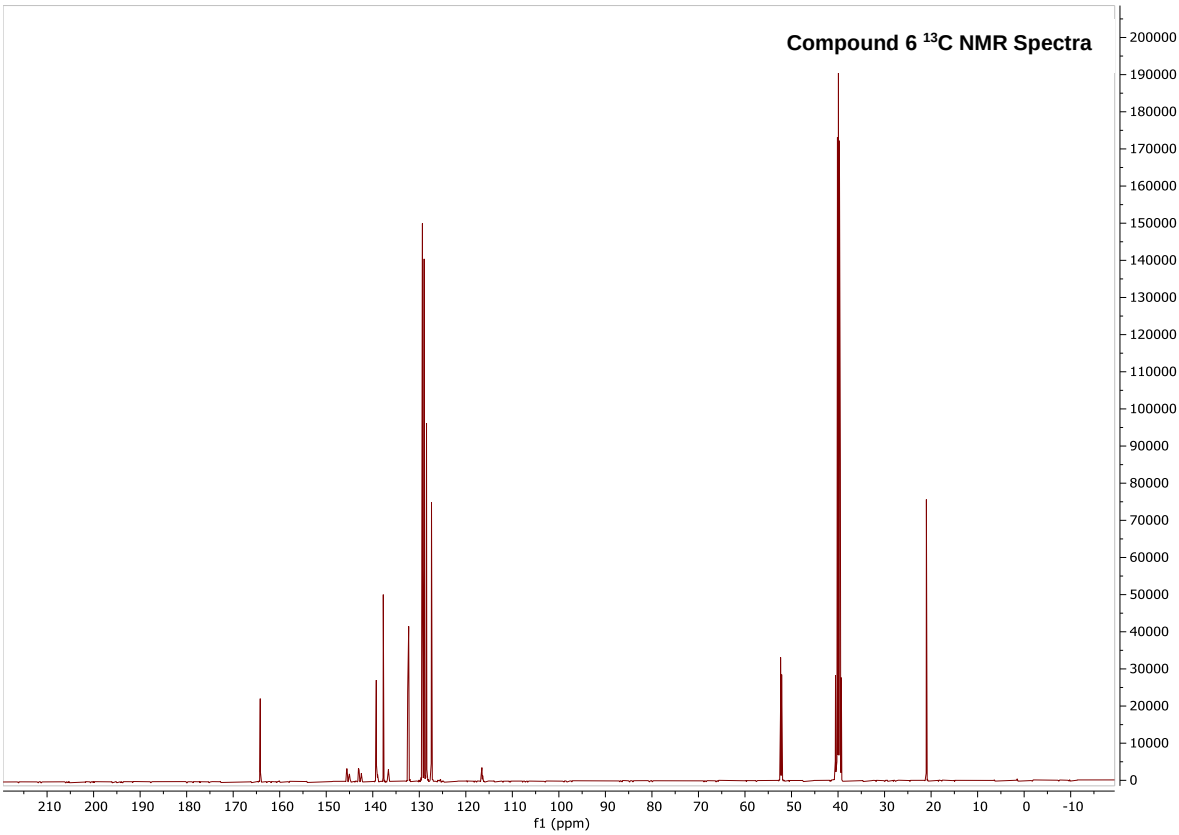
^{1}H NMR (400 MHz, DMSO) δ 11.22 (s, 1H), 9.07 (s, 1H), 7.71 – 7.63 (m, 2H), 7.31 – 7.23 (m, 2H), 7.13 – 7.02 (m, 4H), 4.59 (s, 2H), 4.48 (s, 2H), 2.24 (s, 3H).

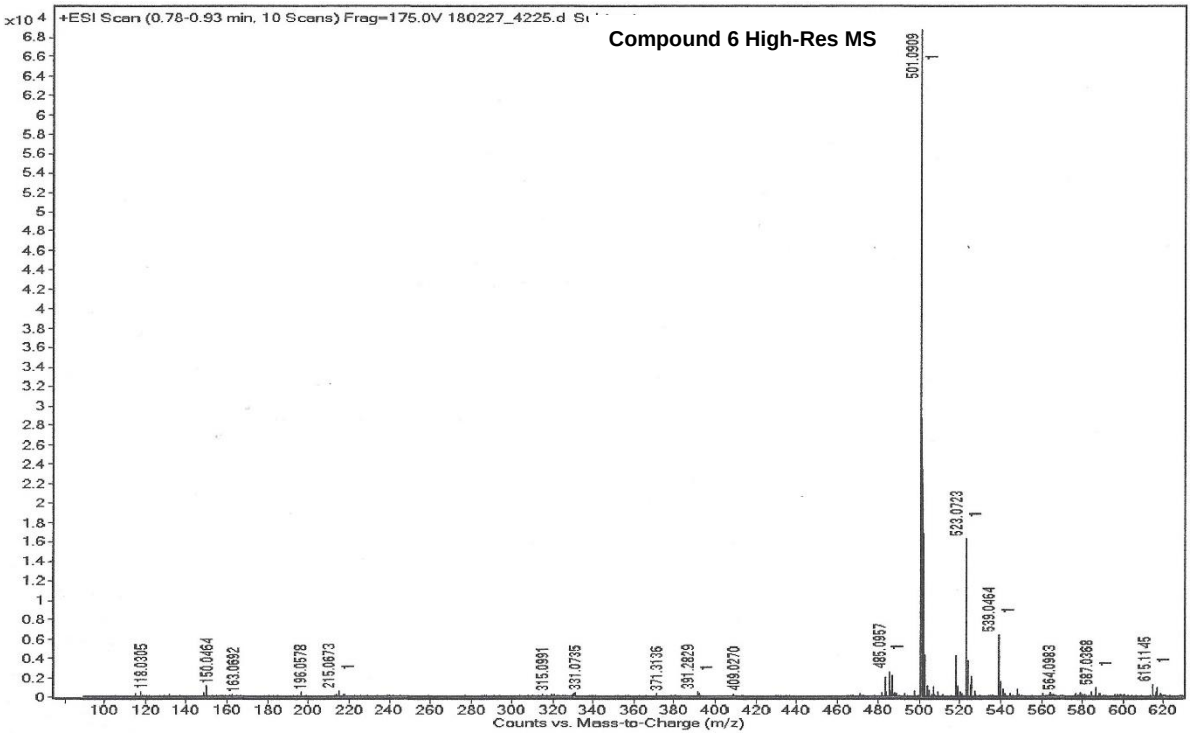
^{19}F NMR (376 MHz, DMSO) δ -135.5 – -138.2 (m), -147.4 (tt, *J* = 22.5, 6.3 Hz), -159.9 (tt, *J* = 22.5, 5.8 Hz).

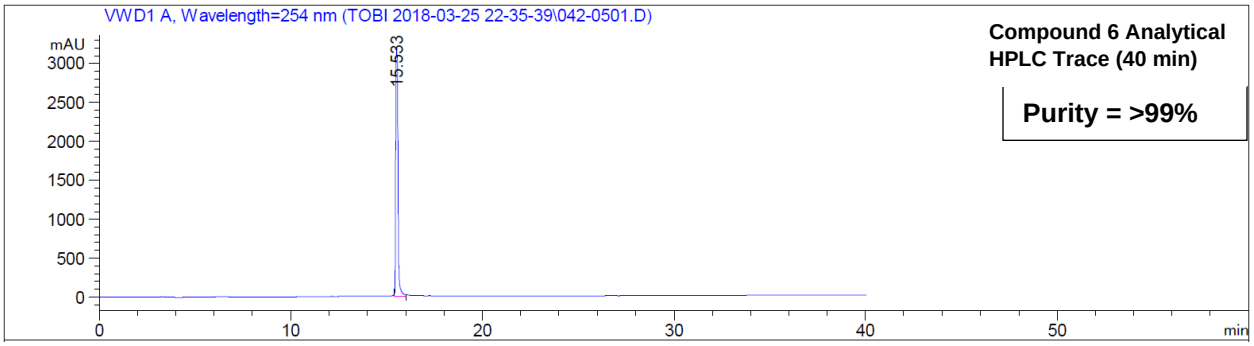
^{13}C NMR (101 MHz, DMSO) δ 164.2, 145.7, 145.6, 145.5, 145.5, 143.1, 143.1, 143.0, 142.9, 139.2, 139.2, 139.1, 137.7, 136.7, 136.5, 132.5, 132.3, 129.3, 129.0, 128.5, 127.4, 116.7, 116.5, 116.4, 52.3, 52.1, 21.0.

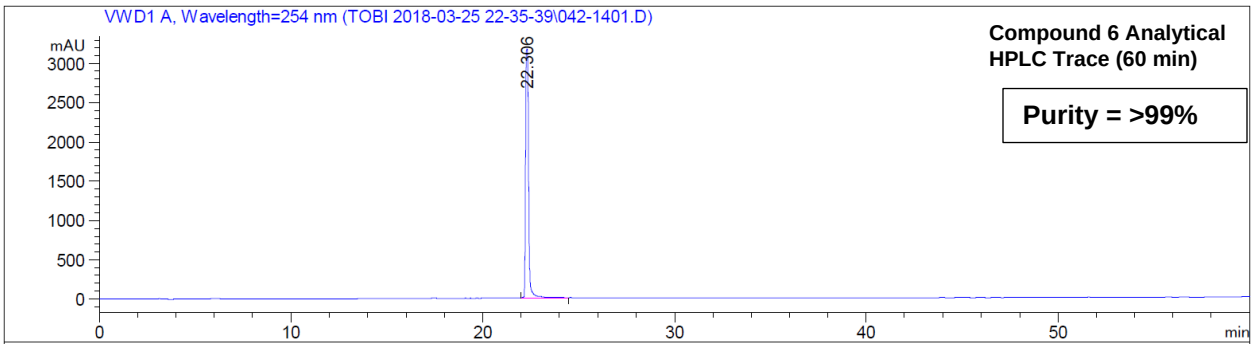


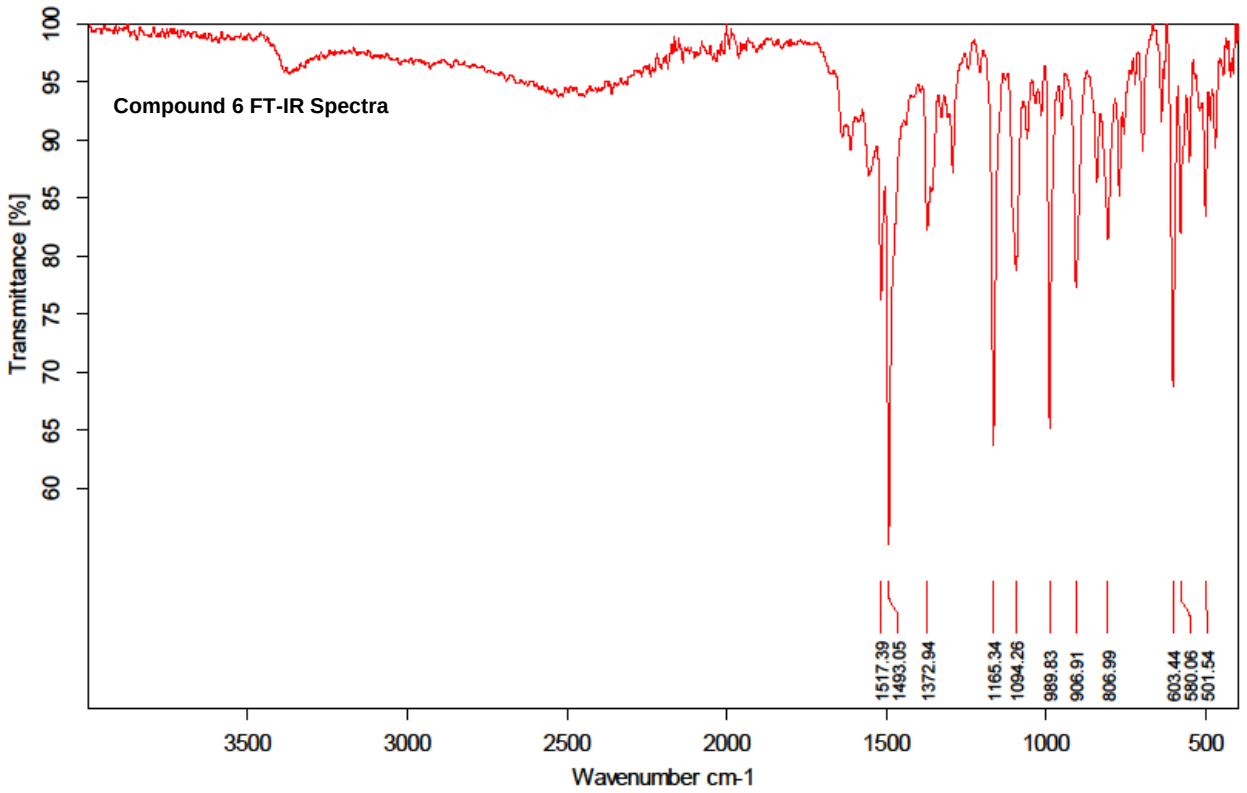

Compound 6 19F NMR Spectra
2.03
1.00
2.11
f1 (ppm)


Compound 6 13C NMR Spectra
f1 (ppm)


Compound 6 High-Res MS
+ESI Scan (0.78-0.93 min, 10 Scans) Frag=175.0V 180227_4225.d
x10 4
Counts vs. Mass-to-Charge (m/z)
118.0305
150.0464
163.0692
196.0578
215.0673
315.0991
331.0735
371.3136
391.2829
409.0270
485.0957
501.0909
523.0723
539.0464
564.0983
587.0368
615.1145


VWD1 A, Wavelength=254 nm (TOBI 2018-03-25 22-35-39\042-0501.D)
mAU
15.533
min
Compound 6 Analytical HPLC Trace (40 min)
Purity = >99%


VWD1 A, Wavelength=254 nm (TOBI 2018-03-25 22-35-39\042-1401.D)
mAU
22.306
min
Compound 6 Analytical HPLC Trace (60 min)
Purity = >99%

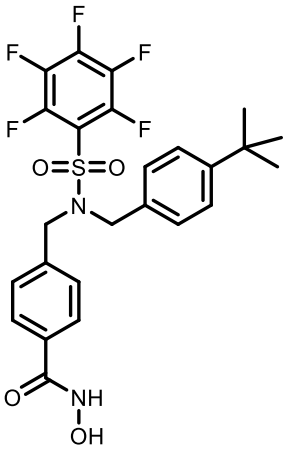


4-(((N-(4-(tert-butyl)benzyl)-2,3,4,5,6-pentafluorophenyl)sulfonamido)methyl)-N-hydroxybenzamide.
Yield: 58%

Compound 7

$C_{25}H_{23}F_5N_2O_4S$

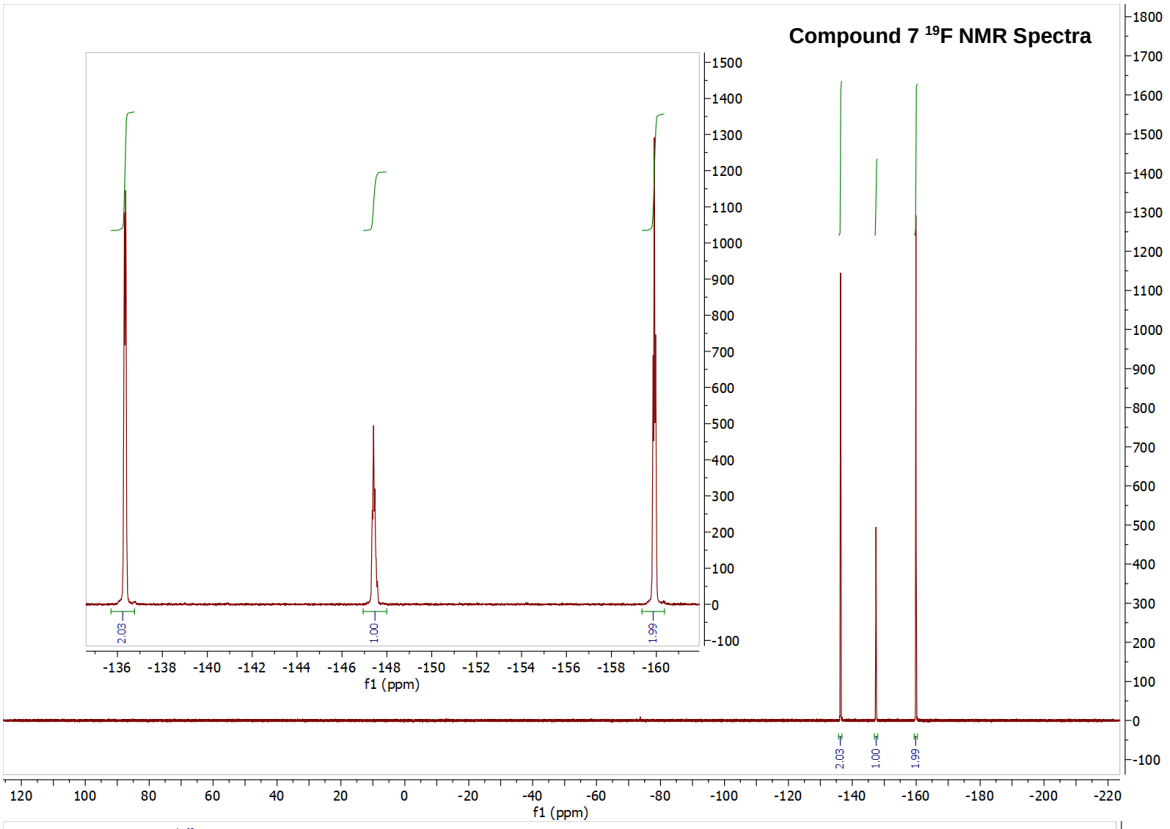
Mol. Weight: 542.53 $gmol^{-1}$

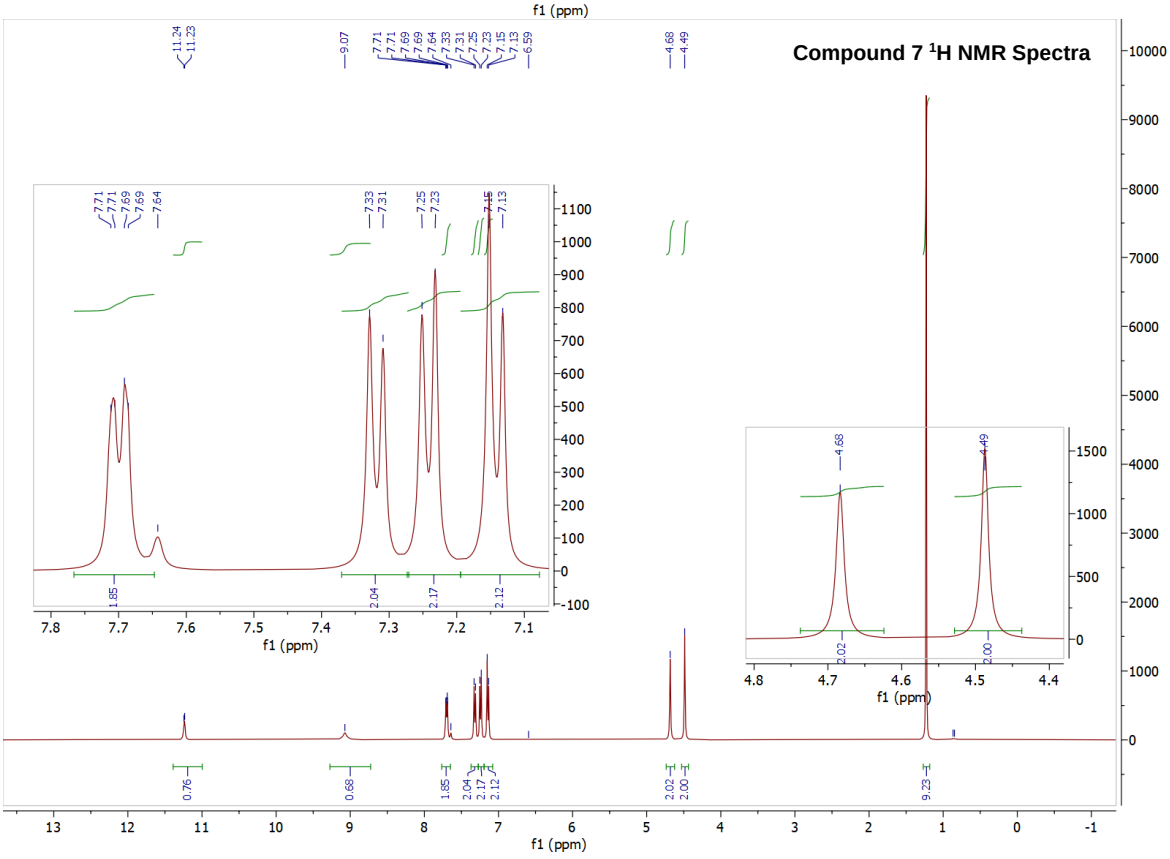


^{1}H NMR (400 MHz, DMSO) δ 11.39 – 11.00 (m, 1H), 9.07 (s, 1H), 7.70 (dd, *J* = 8.0, 2.3 Hz, 2H), 7.32 (d, *J* = 7.9 Hz, 2H), 7.24 (d, *J* = 7.8 Hz, 2H), 7.14 (d, *J* = 8.1 Hz, 2H), 4.68 (s, 2H), 4.49 (s, 2H), 1.23 (s, 9H).

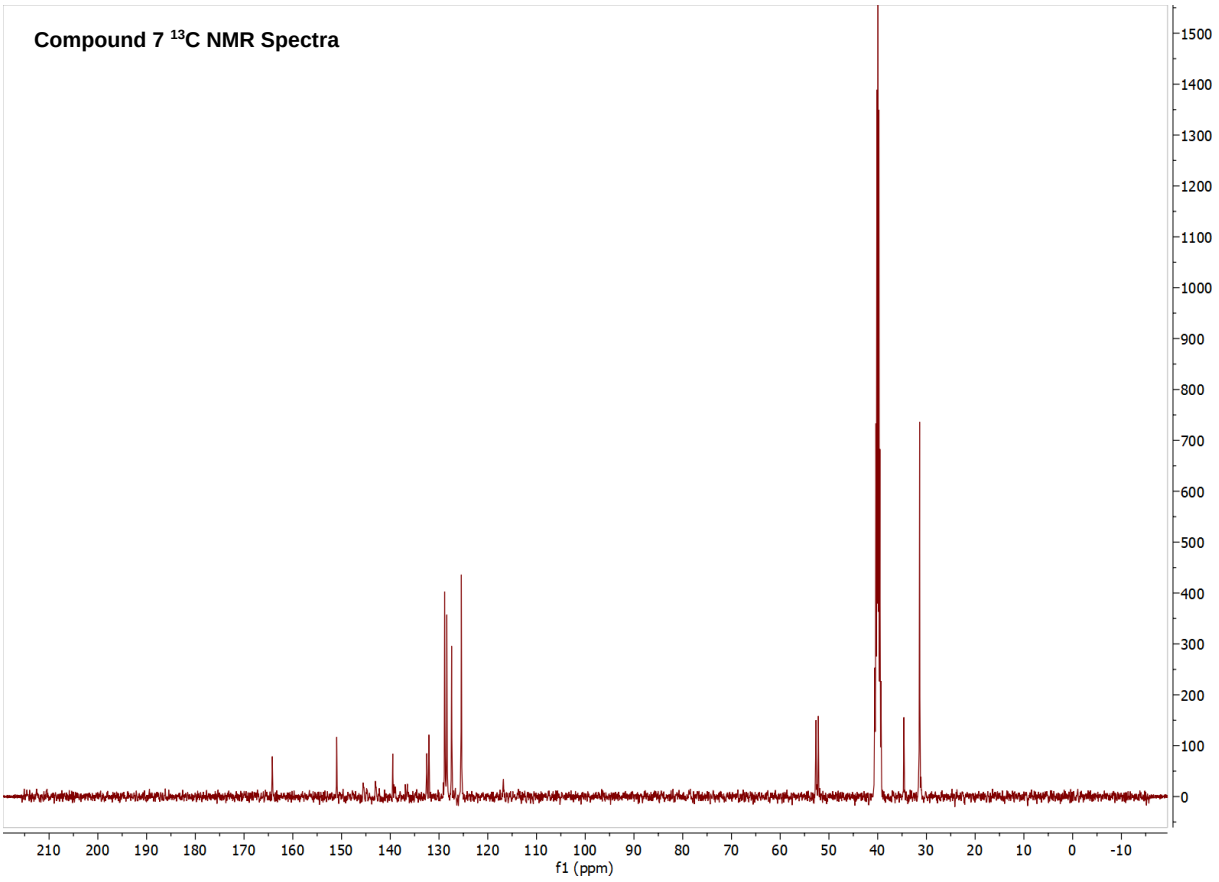
^{19}F NMR (376 MHz, DMSO) δ -136.3 (d, *J* = 23.5 Hz), -147.0 – -148.0 (m), -159.9 (t, *J* = 21.5 Hz).

^{13}C NMR (101 MHz, DMSO) δ 164.2, 151.0, 143.0, 139.5, 132.5, 132.1, 128.8, 128.6, 128.4, 127.4, 125.4, 116.8, 52.7, 52.2, 34.6, 31.4.

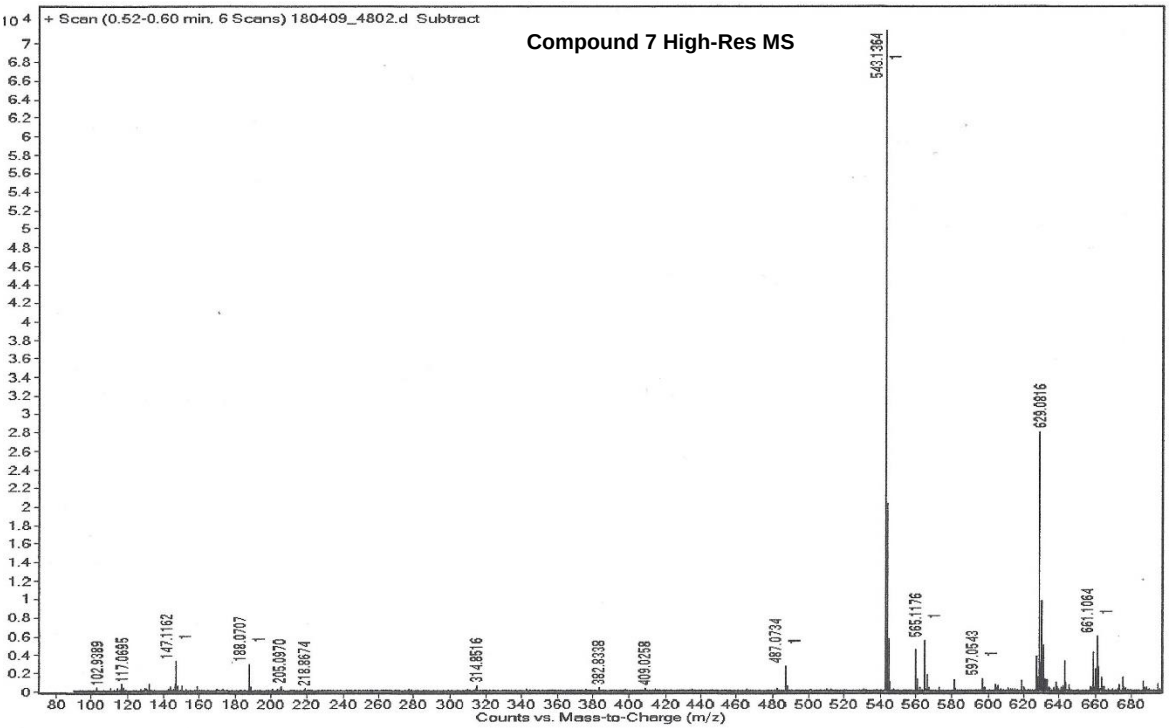

Compound 7 19F NMR Spectra
f1 (ppm)

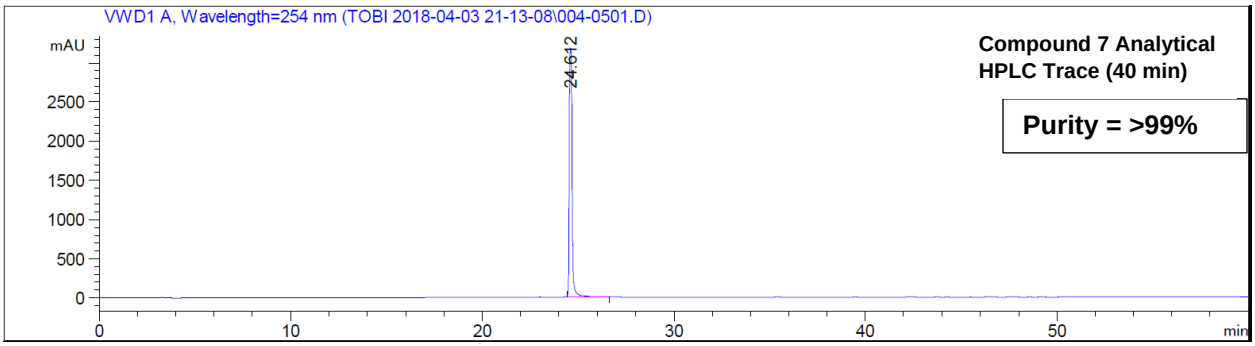

Compound 7 1H NMR Spectra
f1 (ppm)

Compound 7 ^{13}C NMR Spectra



Compound 7 High-Res MS



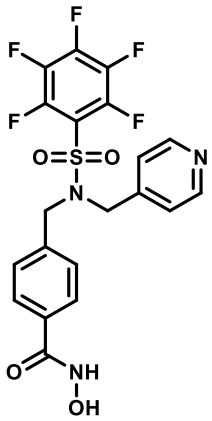


N-hydroxy-4-(((2,3,4,5,6-pentafluoro-N-(pyridin-4-ylmethyl)phenyl)sulfonamido)methyl)benzamide: Yield 39%

Compound 8

$C_{20}H_{14}F_5N_3O_4S$

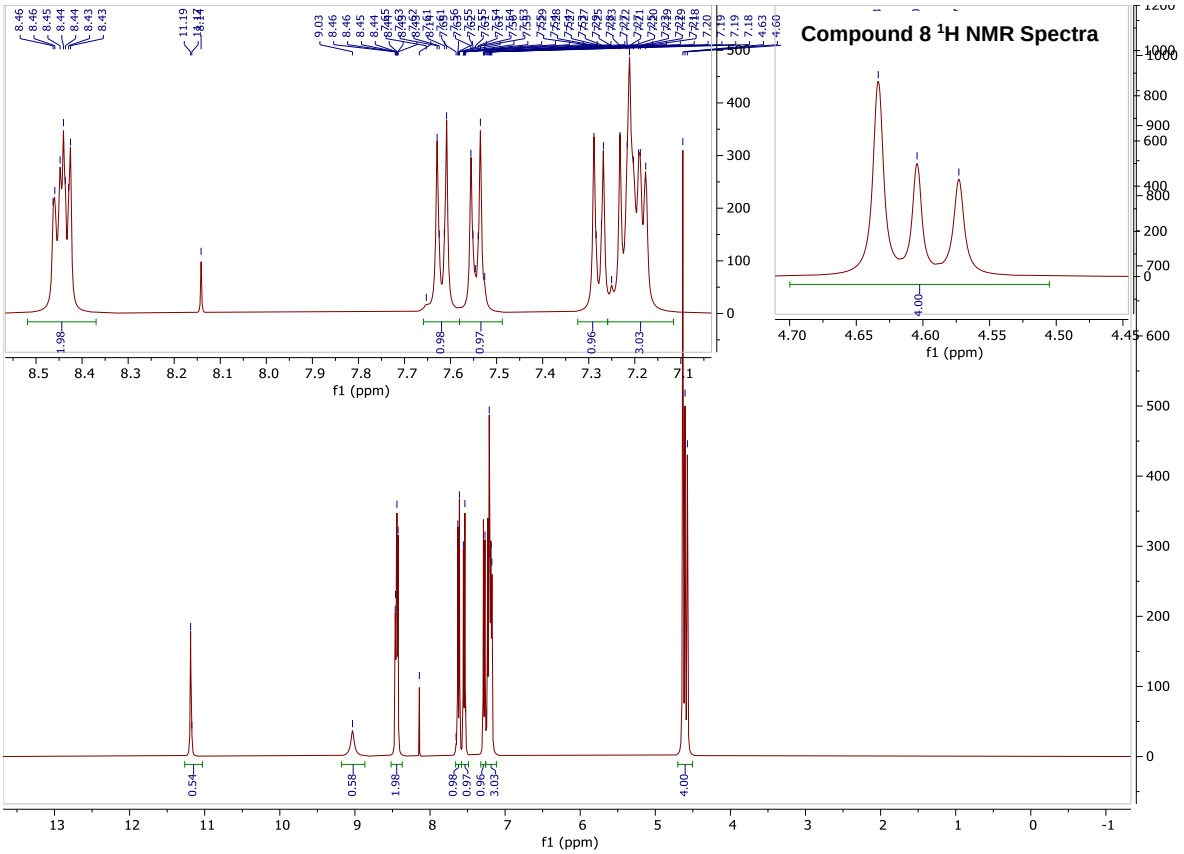
Mol. Weight: 487.41 $gmol^{-1}$

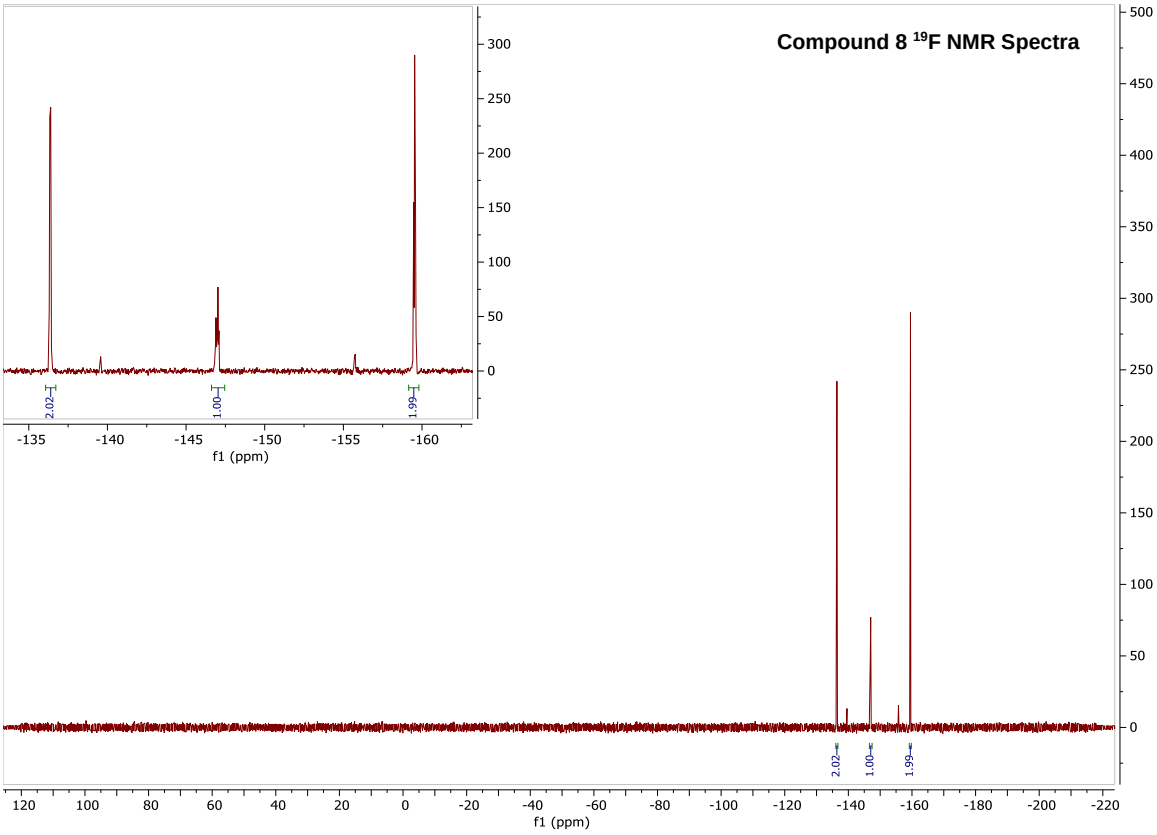


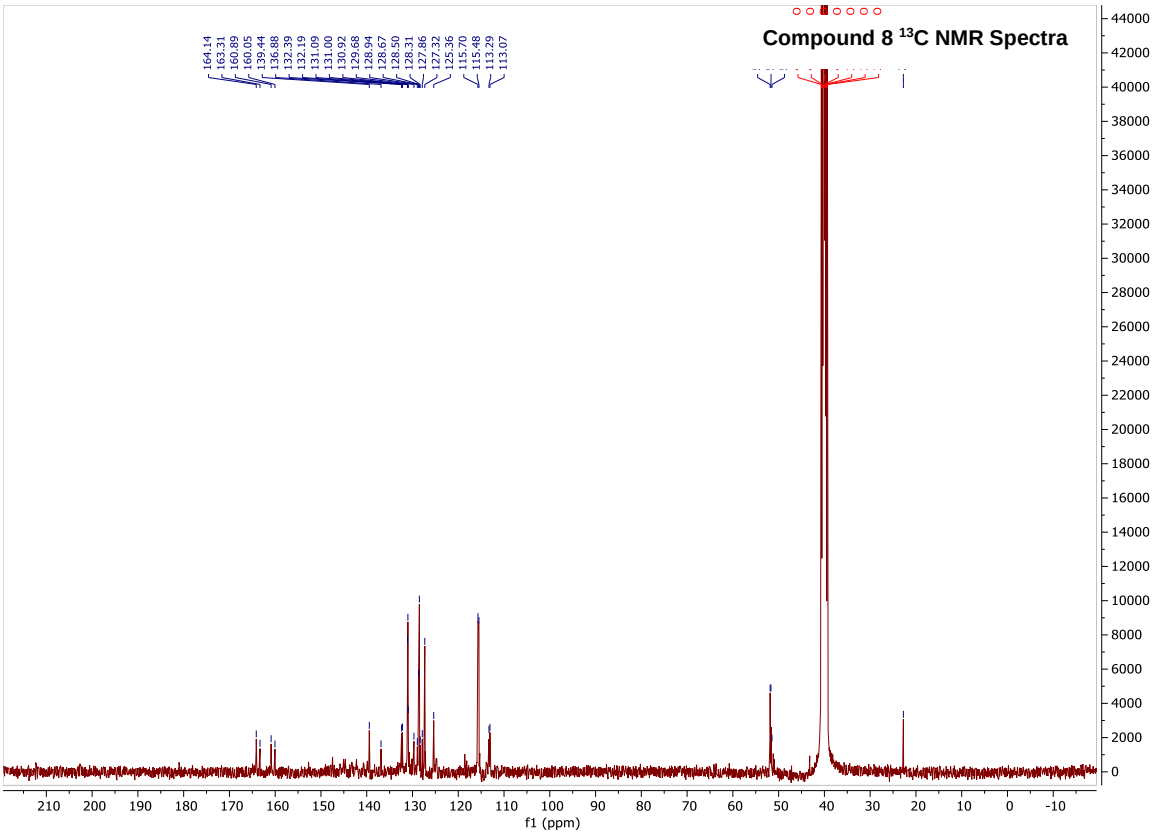
^{1}H NMR (400 MHz, DMSO) δ 11.19 (s, 1H), 9.03 (s, 1H), 8.52 – 8.37 (m, 2H), 7.66 – 7.58 (m, 1H), 7.58 – 7.49 (m, 1H), 7.32 – 7.26 (m, 1H), 7.26 – 7.12 (m, 3H), 4.70 – 4.51 (m, 4H).

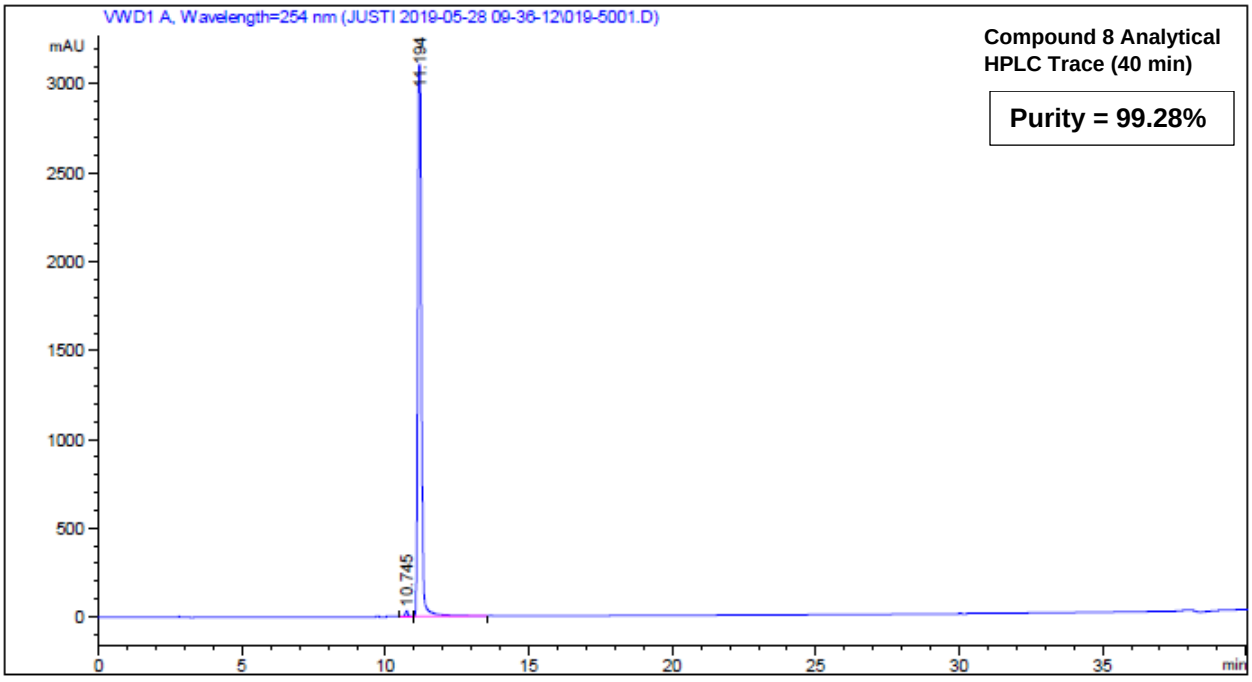
^{19}F NMR (376 MHz, DMSO) δ -136.1 – -136.7 (m), -147.0 (dt, *J* = 46.2, 22.7 Hz), -159.3 – -159.8 (m).

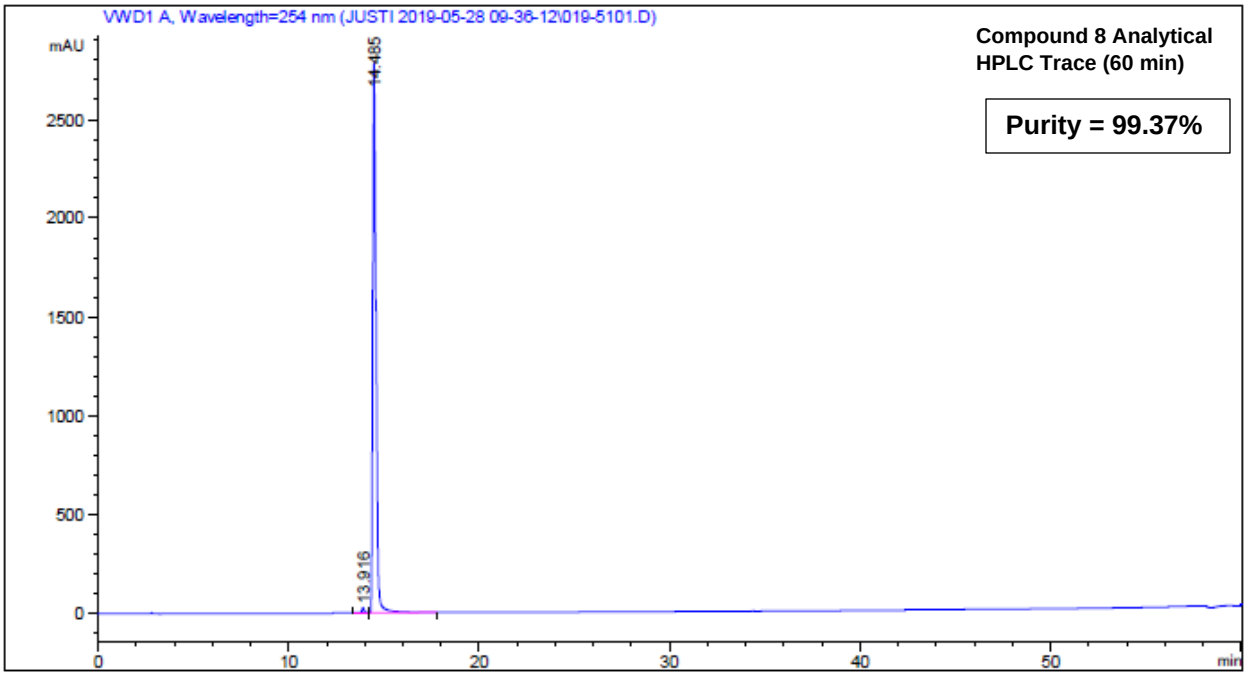
^{13}C NMR (101 MHz, DMSO) δ 164.1, 163.3, 160.9, 160.1, 139.4, 136.9, 132.4, 131.1, 131.0, 129.7, 128.9, 128.7, 128.5, 128.3, 127.9, 127.3, 125.4, 115.7, 113.3, 51.7.

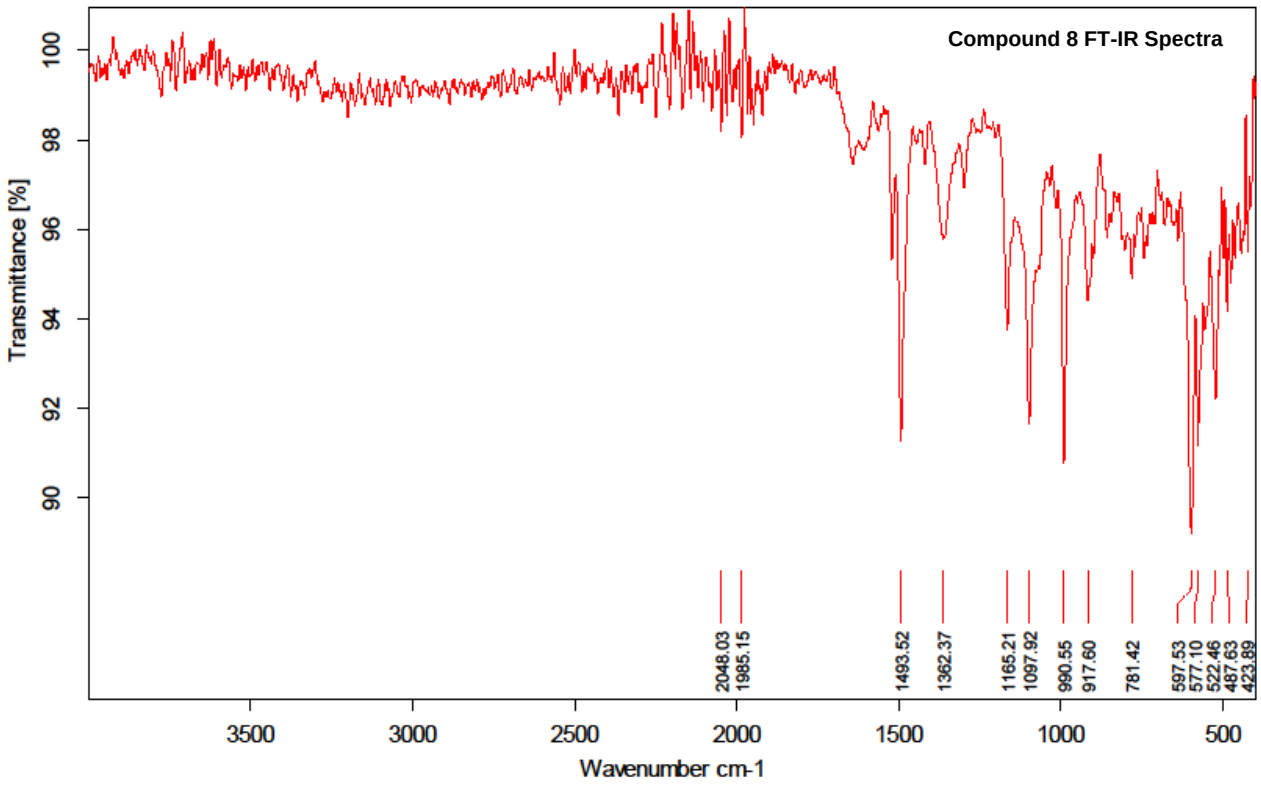

Compound 8 ^{1}H NMR Spectra
f1 (ppm)


Compound 8 ^{19}F NMR Spectra
f1 (ppm)

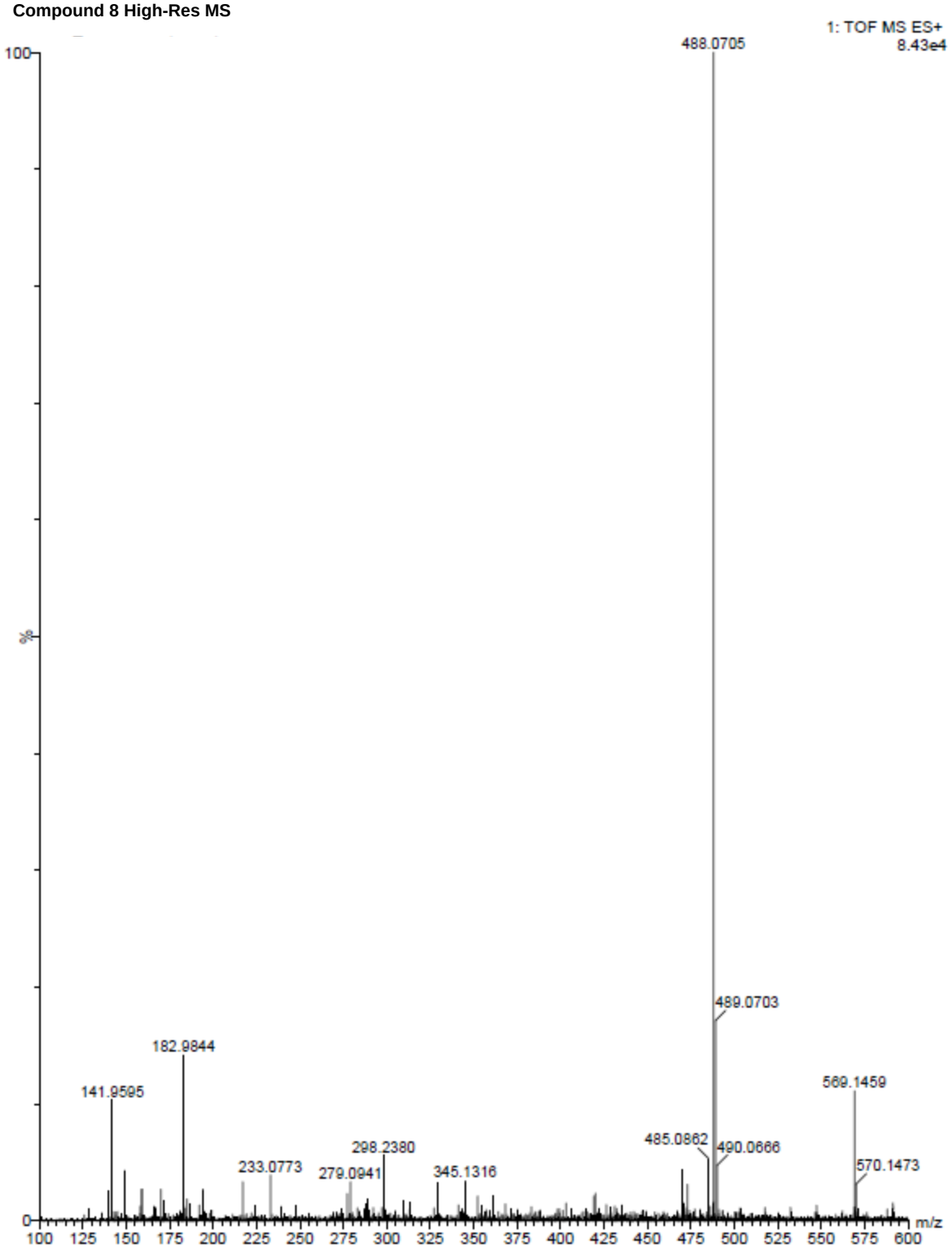

Compound 8 ^{13}C NMR Spectra
164.14
163.31
160.89
160.05
139.44
136.88
132.39
132.19
131.09
131.00
130.92
129.68
128.94
128.67
128.50
128.31
127.86
127.32
125.36
115.70
115.48
113.29
113.07
f1 (ppm)


VWD1 A, Wavelength=254 nm (JUSTI 2019-05-28 09-36-12\019-5001.D)
mAU
11.194
10.745
min
Compound 8 Analytical HPLC Trace (40 min)
Purity = 99.28%


VWD1 A, Wavelength=254 nm (JUSTI 2019-05-28 09-36-12\019-5101.D)
mAU
2500
2000
1500
1000
500
0
14.485
13.916
0
10
20
30
40
50
min
Compound 8 Analytical HPLC Trace (60 min)
Purity = 99.37%


Compound 8 FT-IR Spectra
Transmittance [%]
100
98
96
94
92
90
2048.03
1985.15
1493.52
1362.37
1165.21
1097.92
990.55
917.60
781.42
597.53
577.10
522.46
487.63
423.89
3500
3000
2500
2000
1500
1000
500
Wavenumber cm-1

Compound 8 High-Res MS

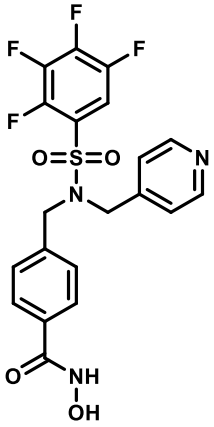


N-hydroxy-4-(((2,3,4,5-tetrafluoro-N-(pyridin-4-ylmethyl)phenyl)sulfonamido)methyl)benzamide

Compound 9

$C_{20}H_{15}F_4N_3O_4S$

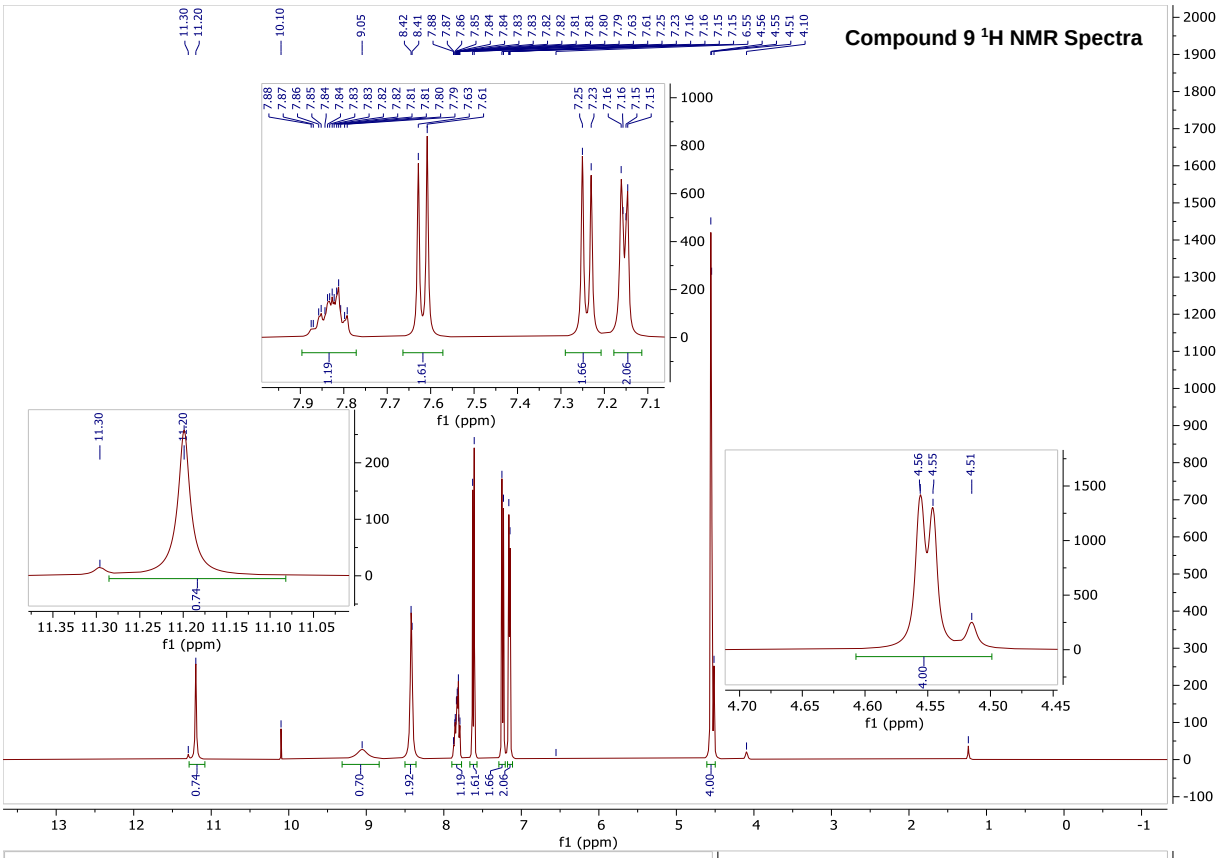
Mol. Weight: 469.42 $gmol^{-1}$

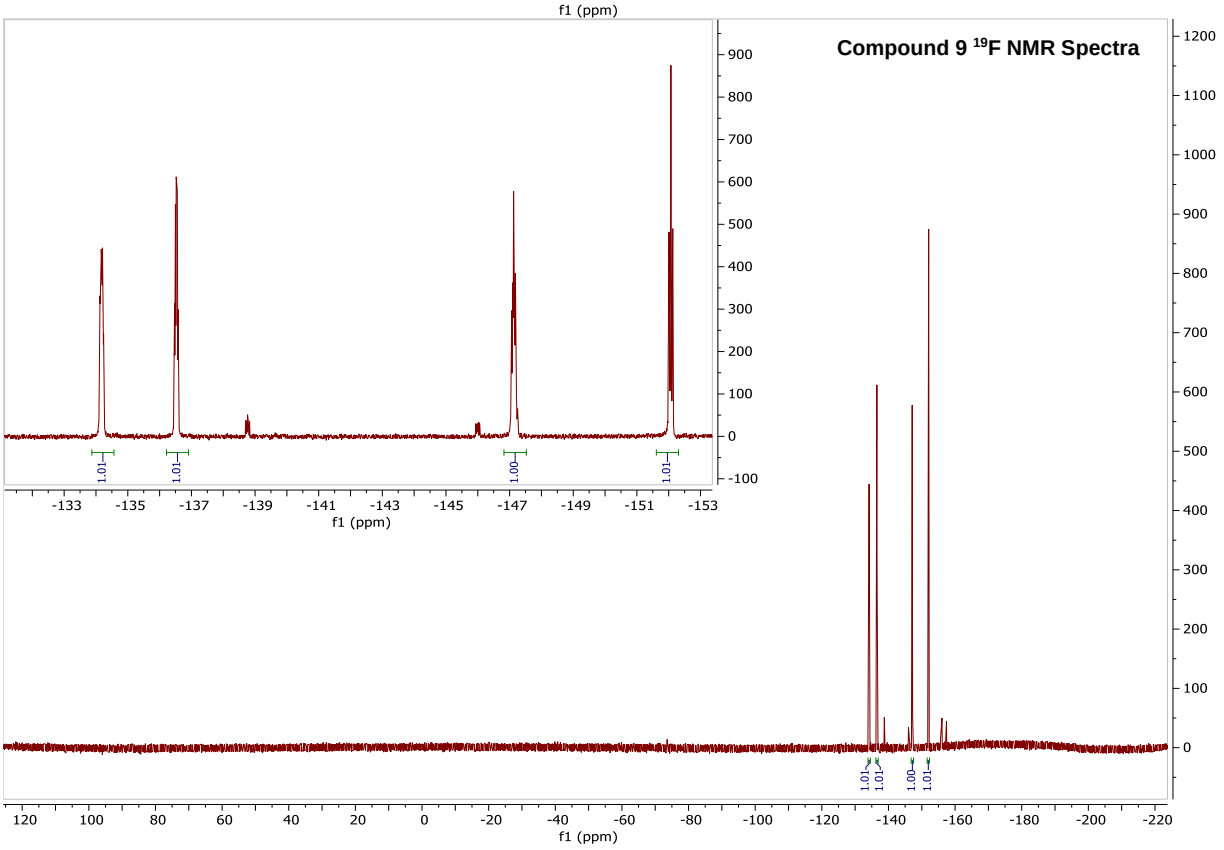


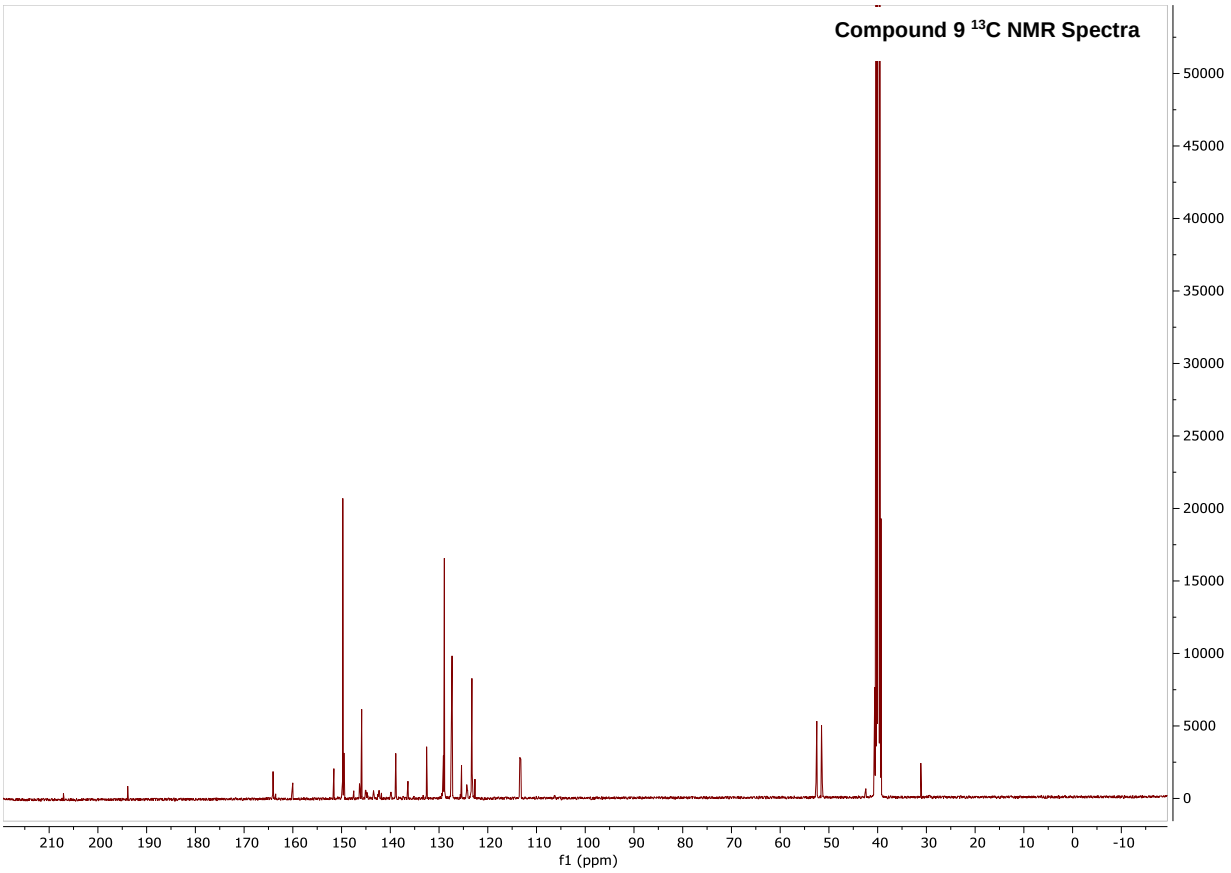
^{1}H NMR (400 MHz, DMSO) δ 11.20 (s, 1H), 9.05 (s, 1H), 8.42 (d, *J* = 5.1 Hz, 2H), 7.82 (dddd, *J* = 9.8, 7.9, 5.9, 2.4 Hz, 1H), 7.62 (d, *J* = 8.2 Hz, 2H), 7.24 (d, *J* = 8.1 Hz, 2H), 7.18 – 7.11 (m, 2H), 4.55 (d, *J* = 4.1 Hz, 4H).

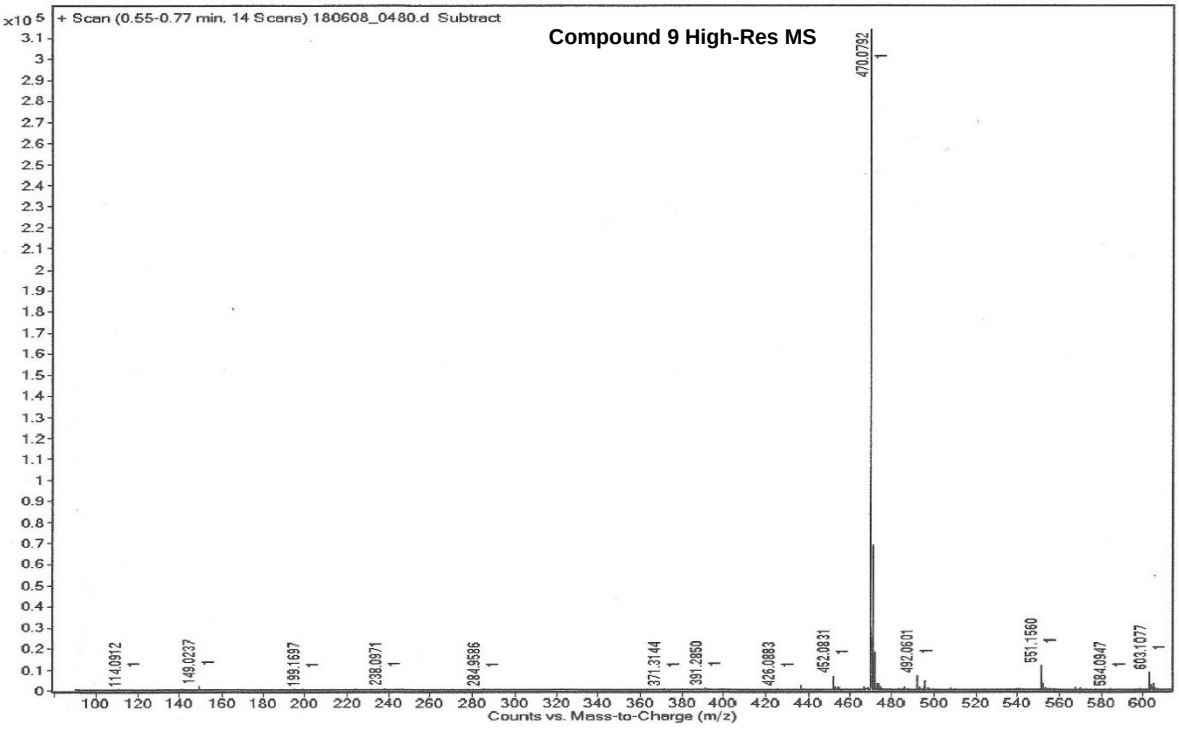
^{19}F NMR (376 MHz, DMSO) δ -134.2 (ddt, *J* = 27.3, 13.4, 7.1 Hz), -136.2 – -136.9 (m), -147.2 (qt, *J* = 25.0, 8.3 Hz), -152.1 (t, *J* = 22.1 Hz).

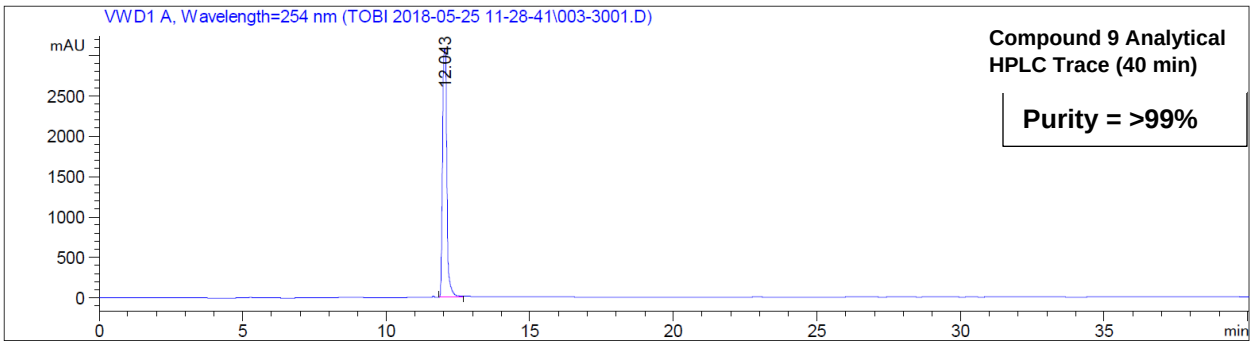
^{13}C NMR (101 MHz, DMSO) δ 193.9, 163.8 (d, *J* = 54.8 Hz), 160.0, 151.6, 149.8, 149.5, 145.9, 138.9, 136.4, 132.6, 129.1, 128.9, 127.3, 125.4, 123.3, 113.3 (dd, *J* = 22.0, 3.4 Hz), 52.6 (d, *J* = 16.3 Hz), 52.1 – 50.1 (m), 31.2.

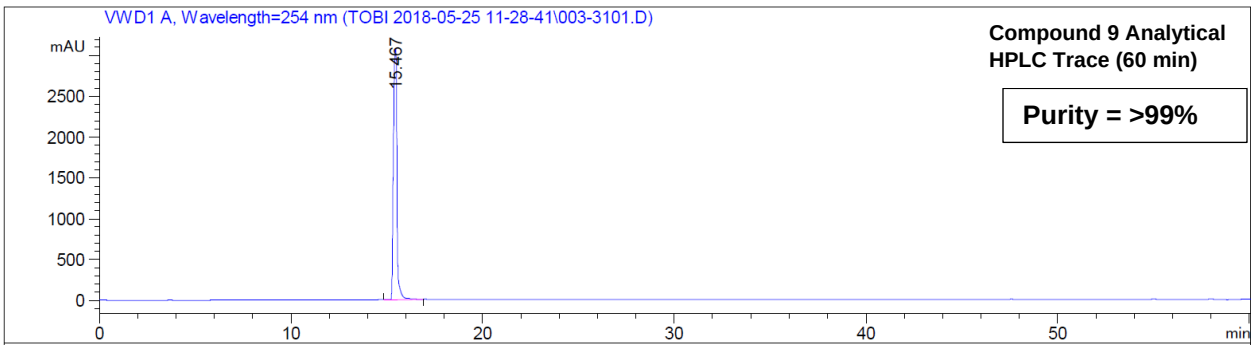

Compound 9 ^{1}H NMR Spectra
f1 (ppm)


Compound 9 ^{19}F NMR Spectra
f1 (ppm)


Compound 9 13C NMR Spectra
f1 (ppm)


+ Scan (0.55-0.77 min, 14 Scans) 180608_0480.d Subtract
Compound 9 High-Res MS
Counts vs. Mass-to-Charge (m/z)



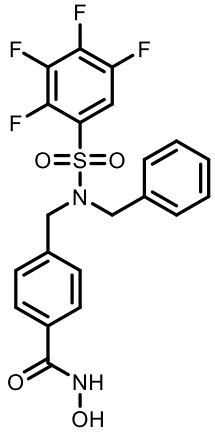


4-(((N-benzyl-2,3,4,5-tetrafluorophenyl)sulfonamido)methyl)-N-hydroxybenzamide. Yield: 65%

Compound 10

$C_{21}H_{16}F_4N_2O_4S$

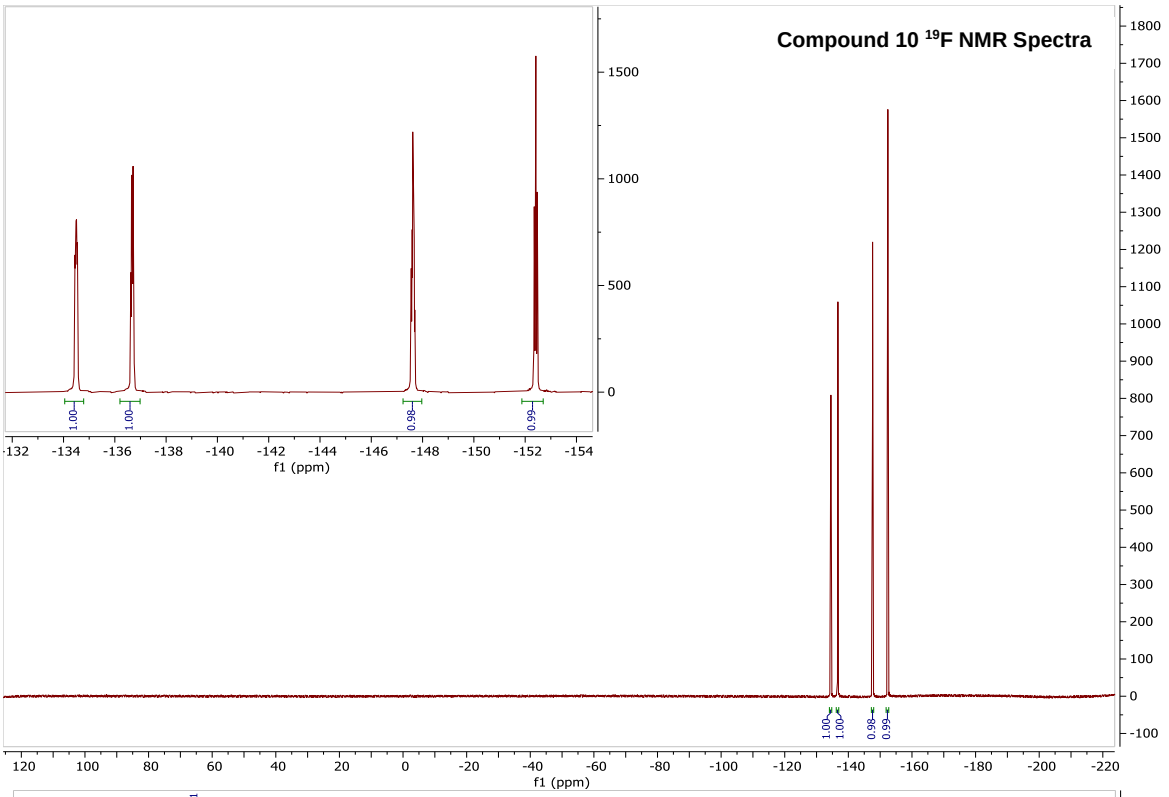
Mol. Weight: 468.42 $gmol^{-1}$

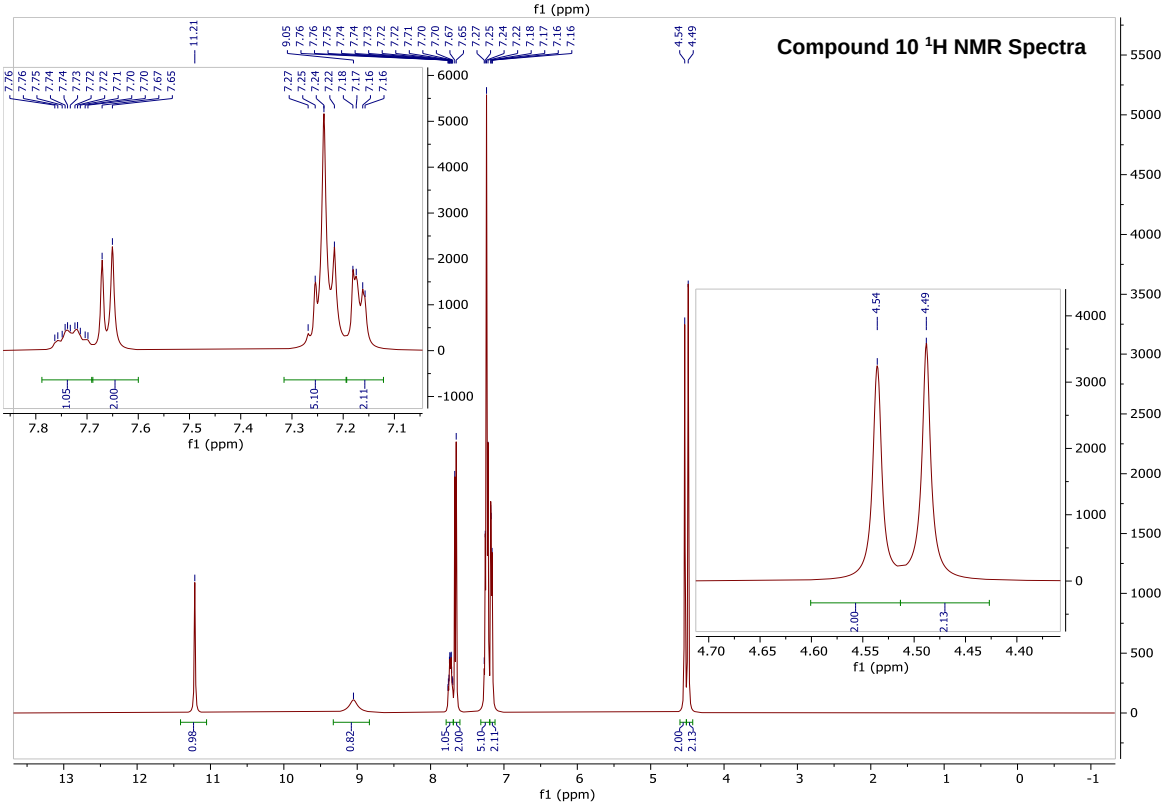


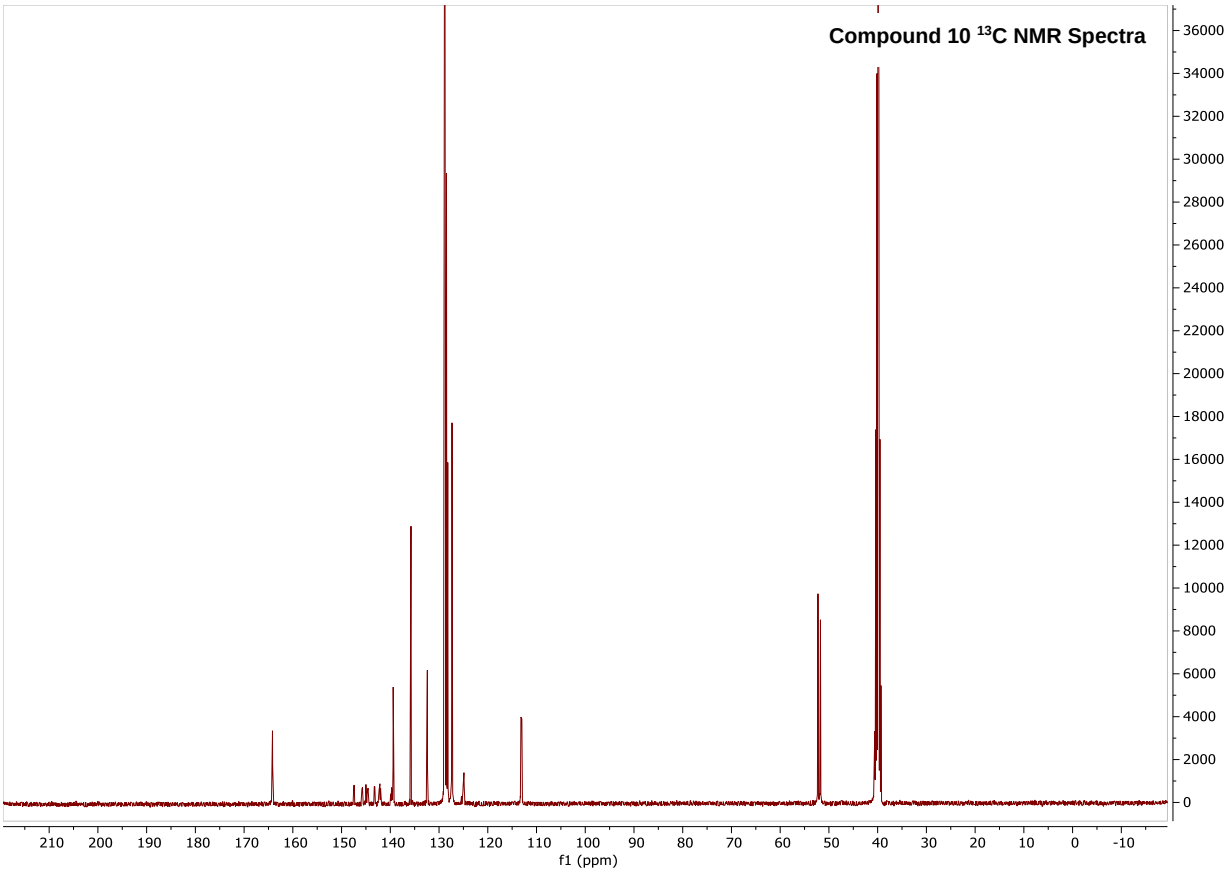
^{1}H NMR (400 MHz, DMSO) δ 11.21 (s, 1H), 9.05 (s, 1H), 7.73 (dddd, *J* = 10.2, 8.1, 5.9, 2.2 Hz, 1H), 7.66 (d, *J* = 8.1 Hz, 2H), 7.32 – 7.19 (m, 5H), 7.17 (dd, *J* = 7.3, 2.2 Hz, 2H), 4.54 (s, 2H), 4.49 (s, 2H).

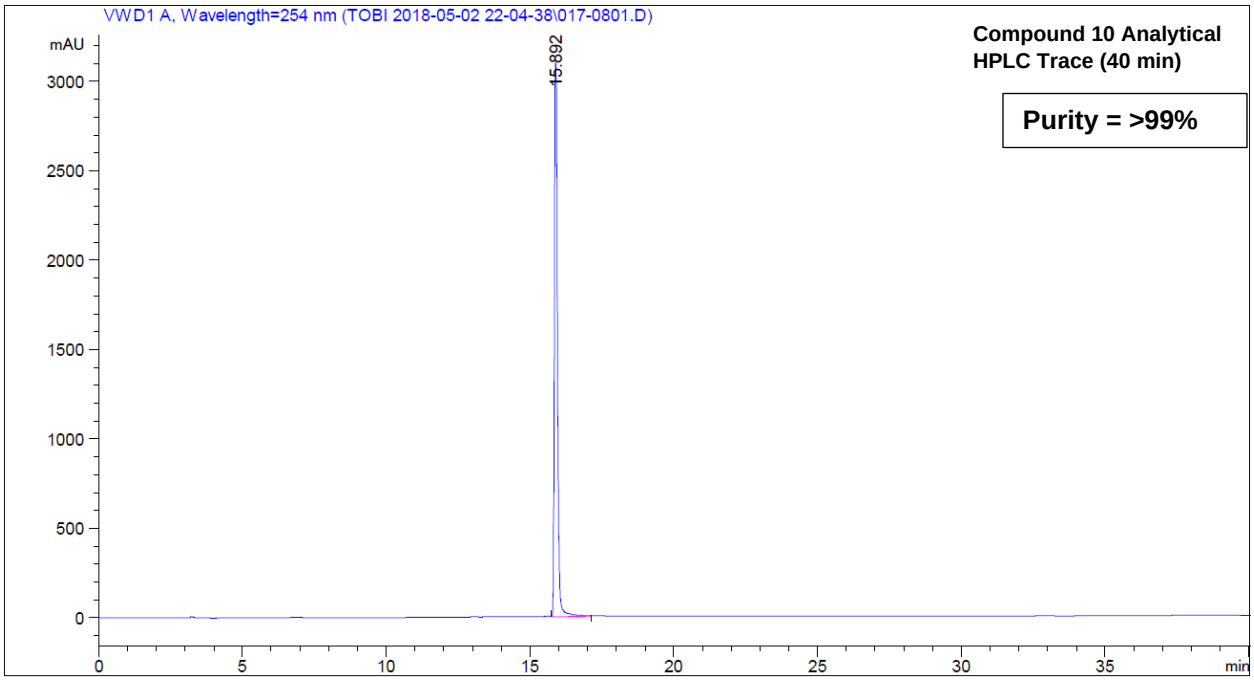
^{19}F NMR (376 MHz, DMSO) δ -134.5 (ddt, *J* = 20.2, 13.6, 7.1 Hz), -136.7 (dt, *J* = 22.2, 11.0 Hz), -147.6 (tt, *J* = 21.7, 7.9 Hz), -152.4 (t, *J* = 22.0 Hz).

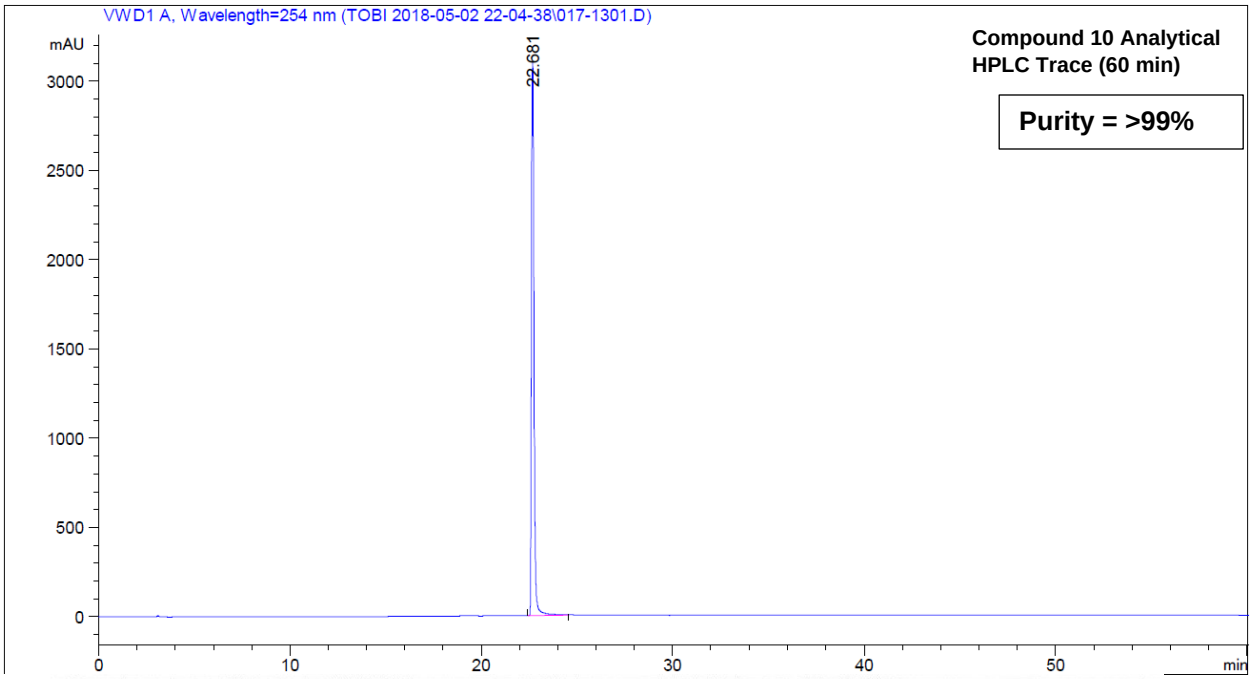
^{13}C NMR (101 MHz, DMSO) δ 164.2, 147.4 (d, *J* = 9.9 Hz), 145.8 (d, *J* = 11.6 Hz), 145.0 (d, *J* = 10.2 Hz), 144.6 (t, *J* = 15.0 Hz), 143.2 (d, *J* = 11.7 Hz), 142.1 (dt, *J* = 27.0, 13.3 Hz), 139.8 (t, *J* = 14.8 Hz), 139.4, 135.8, 132.4, 128.9, 128.8, 128.5, 128.2, 127.3, 113.1 (dd, *J* = 22.0, 3.2 Hz), 52.2 (d, *J* = 1.9 Hz), 51.8 (d, *J* = 2.2 Hz).

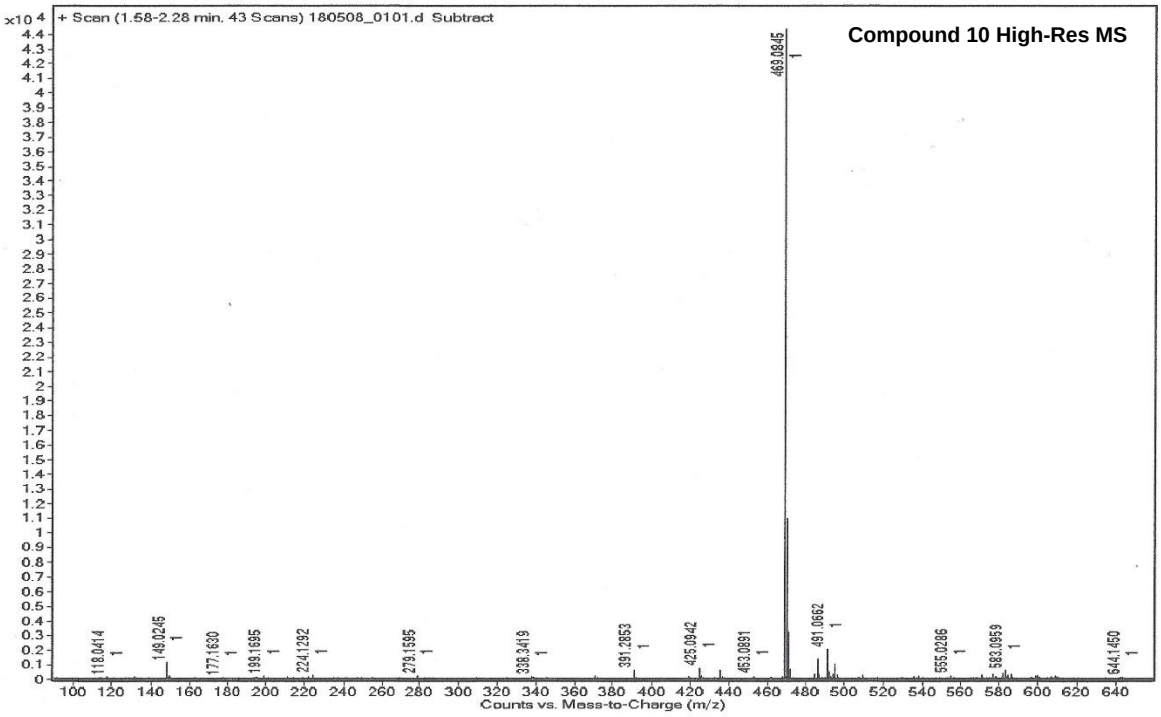

Compound 10 19F NMR Spectra
f1 (ppm)

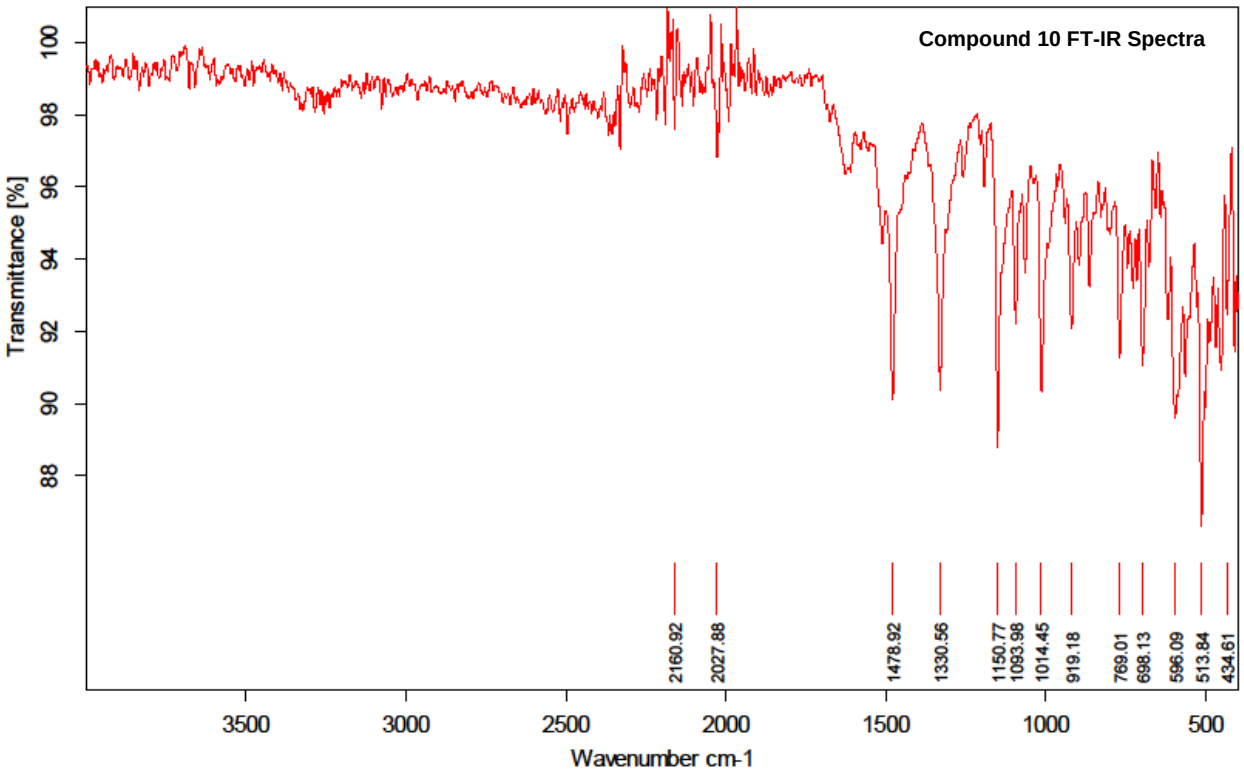

Compound 10 1H NMR Spectra
f1 (ppm)


Compound 10 ^{13}C NMR Spectra
f1 (ppm)


VWD1 A, Wavelength=254 nm (TOBI 2018-05-02 22-04-38\017-0801.D)
mAU
15.892
min
Compound 10 Analytical HPLC Trace (40 min)
Purity = >99%


VWD1 A, Wavelength=254 nm (TOBI 2018-05-02 22-04-38\017-1301.D)
mAU
22.681
min
Compound 10 Analytical HPLC Trace (60 min)
Purity = >99%


+ Scan (1.58-2.28 min, 43 Scans) 180508_0101.d Subtract
x10 4
469.0845
Compound 10 High-Res MS
Counts vs. Mass-to-Charge (m/z)

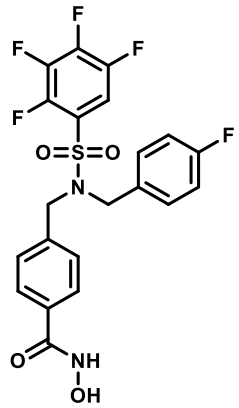


N-hydroxy-4-(((2,3,4,5-tetrafluoro-N-(4-fluorobenzyl)phenyl)sulfonamido)methyl)benzamide. Yield: 59%

Compound 11

$C_{21}H_{15}F_5N_2O_4S$

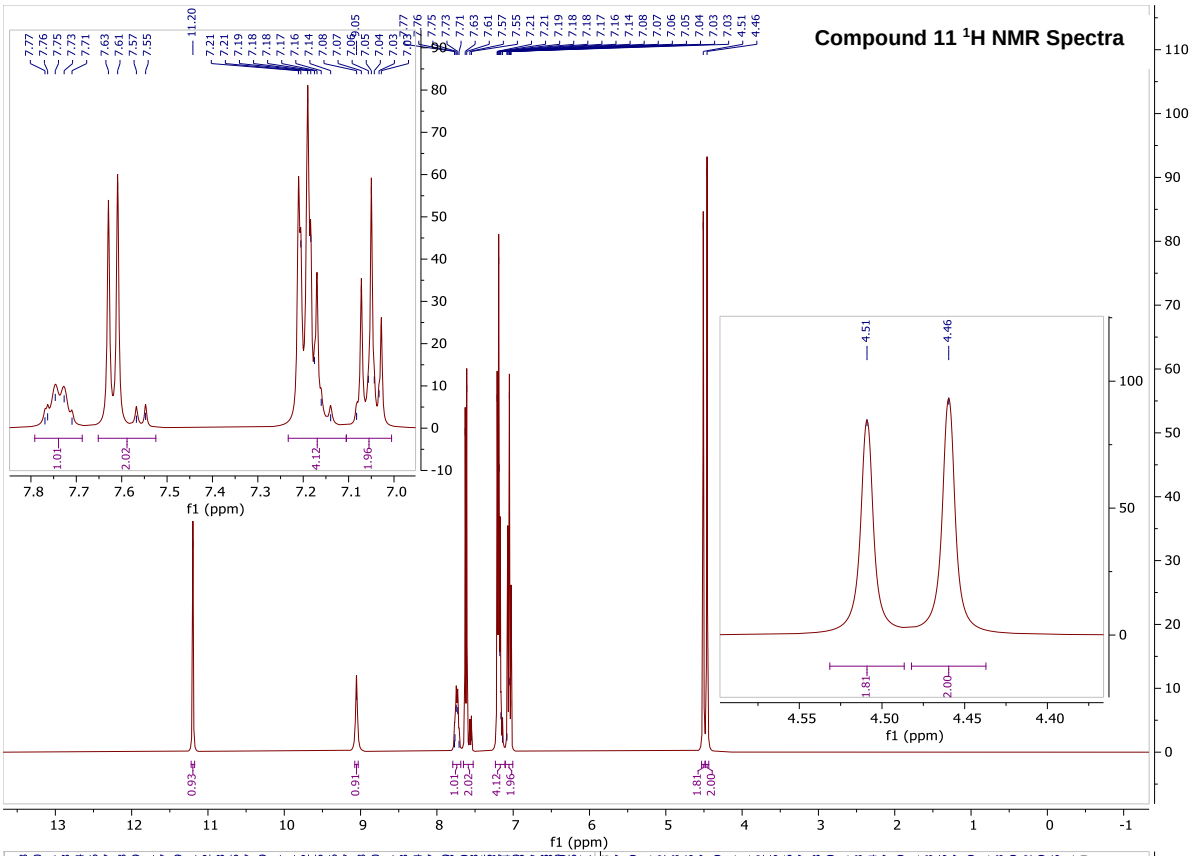
Mol. Weight: 486.41 $gmol^{-1}$

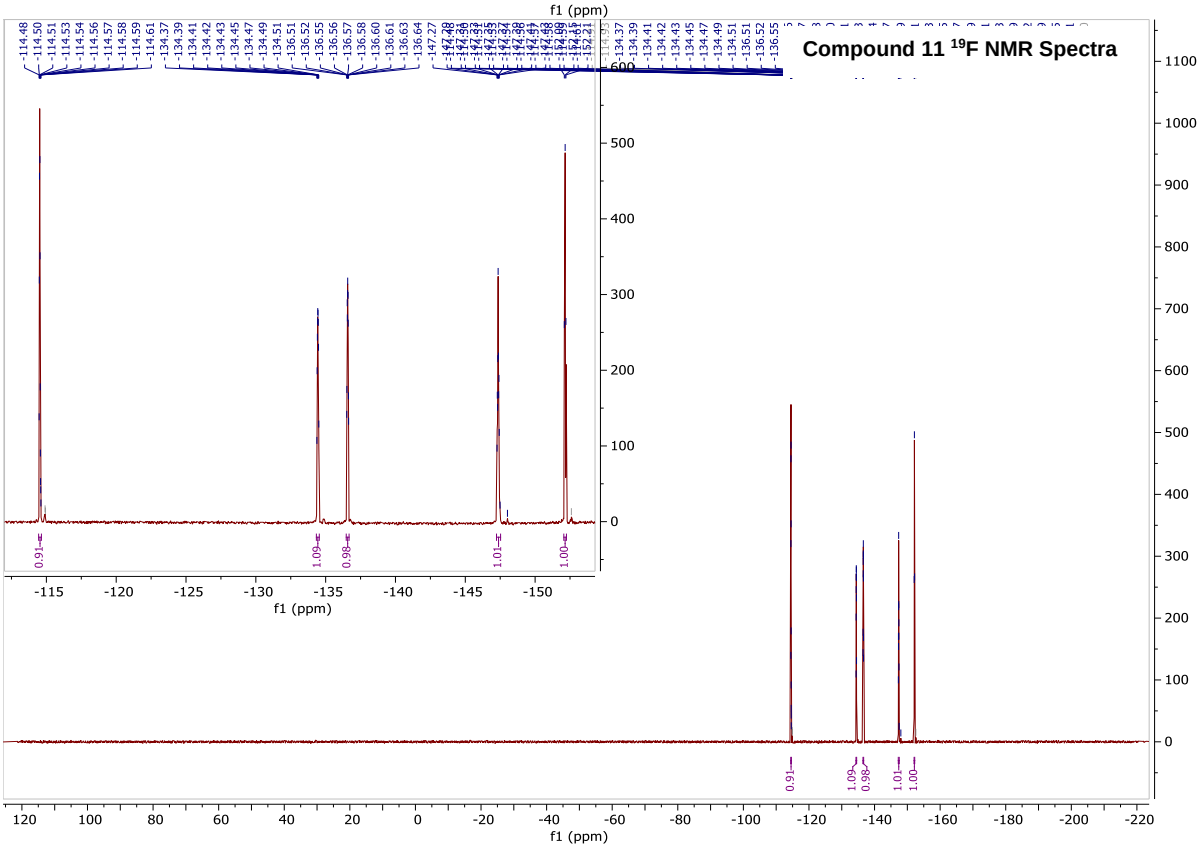


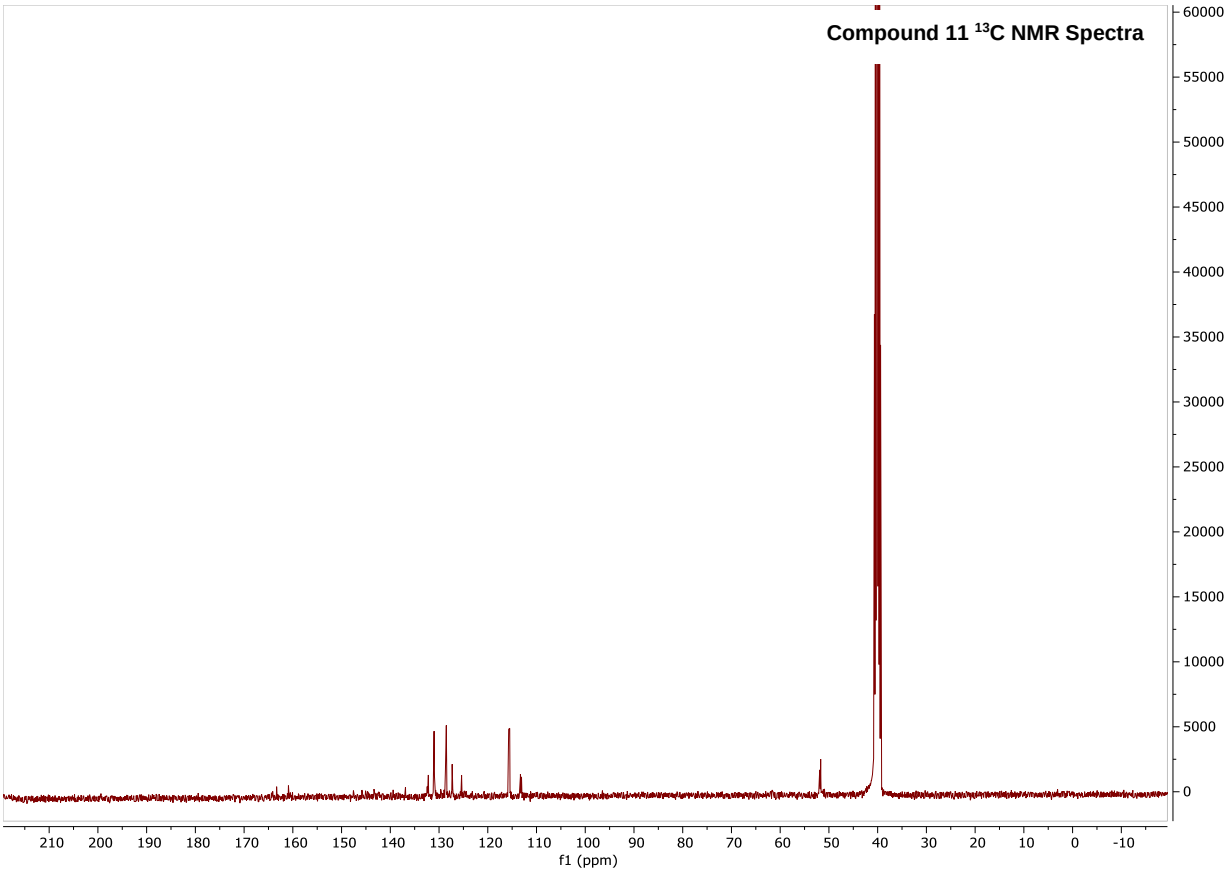
^{1}H NMR (400 MHz, DMSO) δ 11.20 (s, 1H), 9.05 (s, 1H), 7.79 – 7.69 (m, 1H), 7.65 – 7.52 (m, 2H), 7.23 – 7.11 (m, 4H), 7.10 – 7.00 (m, 2H), 4.51 (s, 2H), 4.46 (s, 2H).

^{13}C NMR (101 MHz, DMSO) δ 160.9, 132.2, 131.1, 131.0, 128.7, 128.5, 127.3, 125.4, 115.7, 115.5, 113.3, 113.1, 51.9, 51.7.

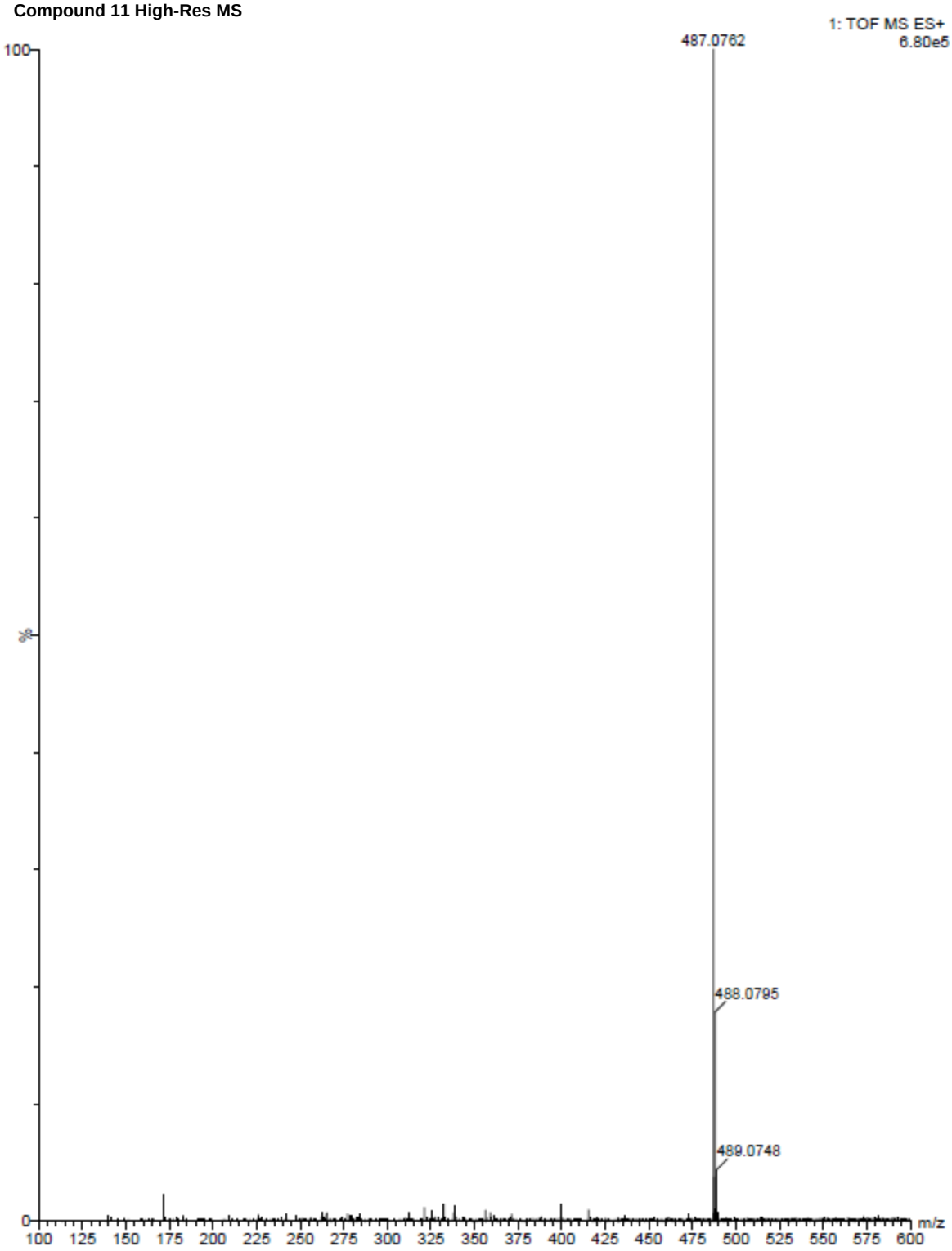
^{19}F NMR (376 MHz, DMSO) δ -114.5 (ddt, *J* = 14.7, 9.1, 5.0 Hz), -134.4 (ddt, *J* = 27.5, 13.5, 7.2 Hz), -136.6 (dtd, *J* = 23.4, 11.7, 10.4, 3.5 Hz), -147.4 (tt, *J* = 21.5, 8.1 Hz), -152.2 (t, *J* = 21.8 Hz).

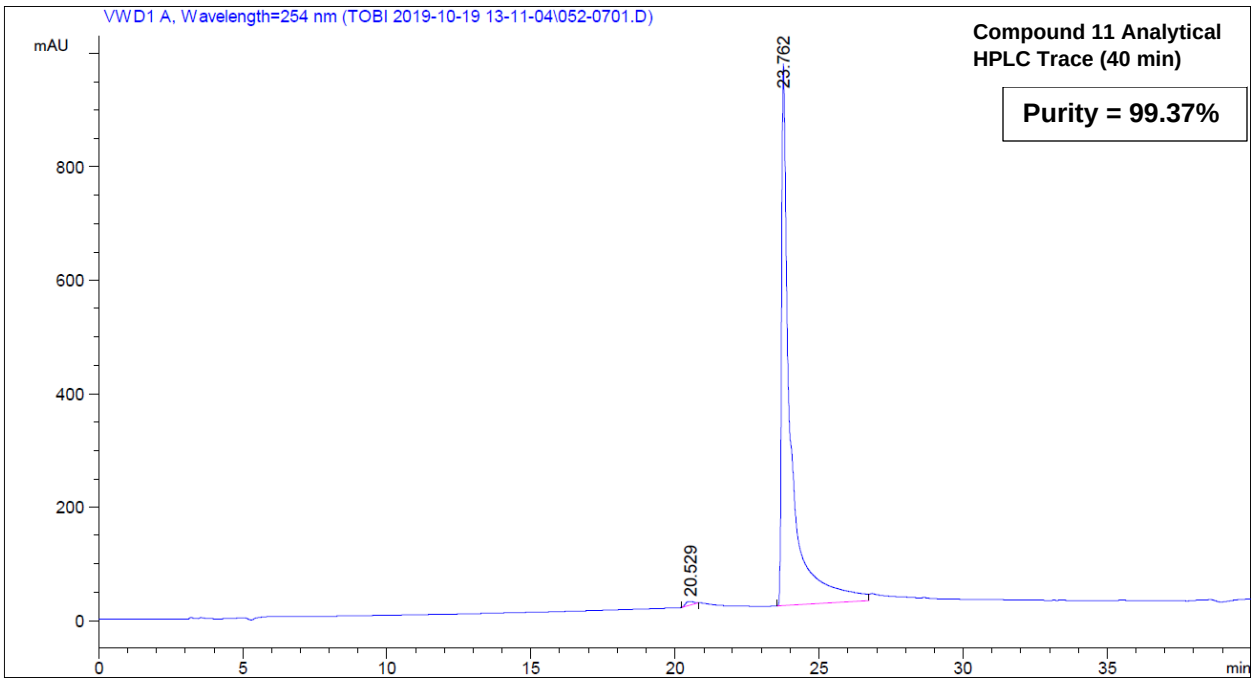

Compound 11 ^{1}H NMR Spectra
f1 (ppm)

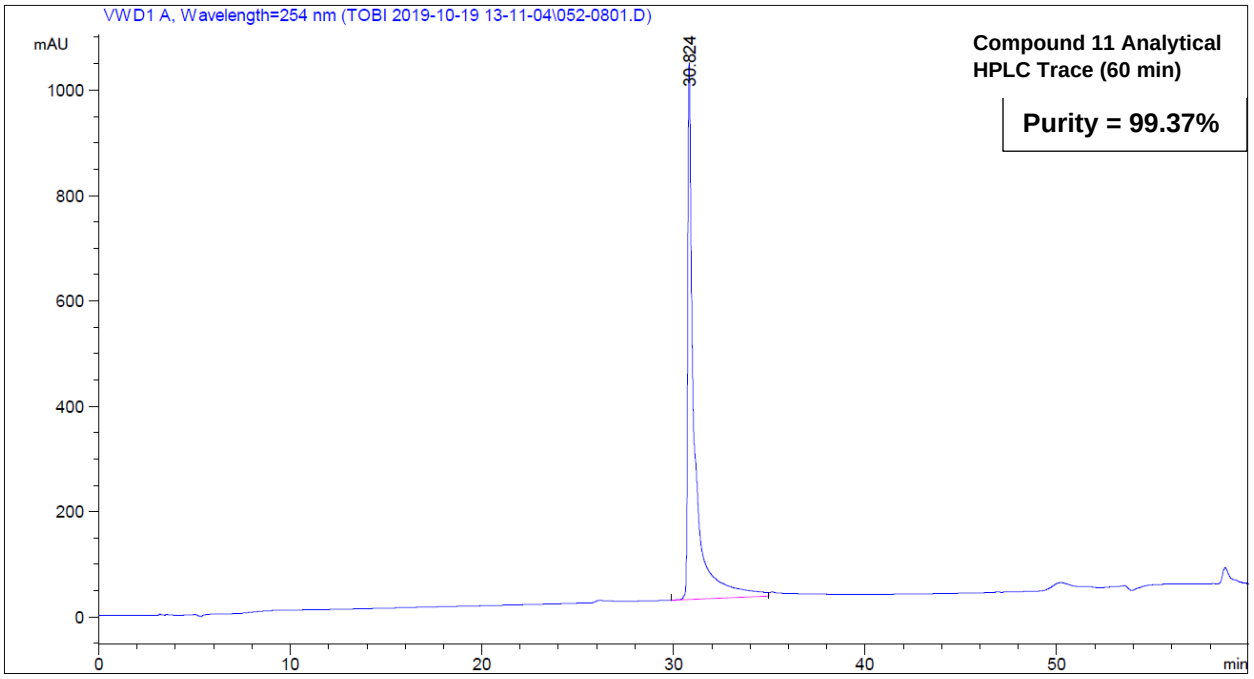

Compound 11 ^{19}F NMR Spectra
f1 (ppm)

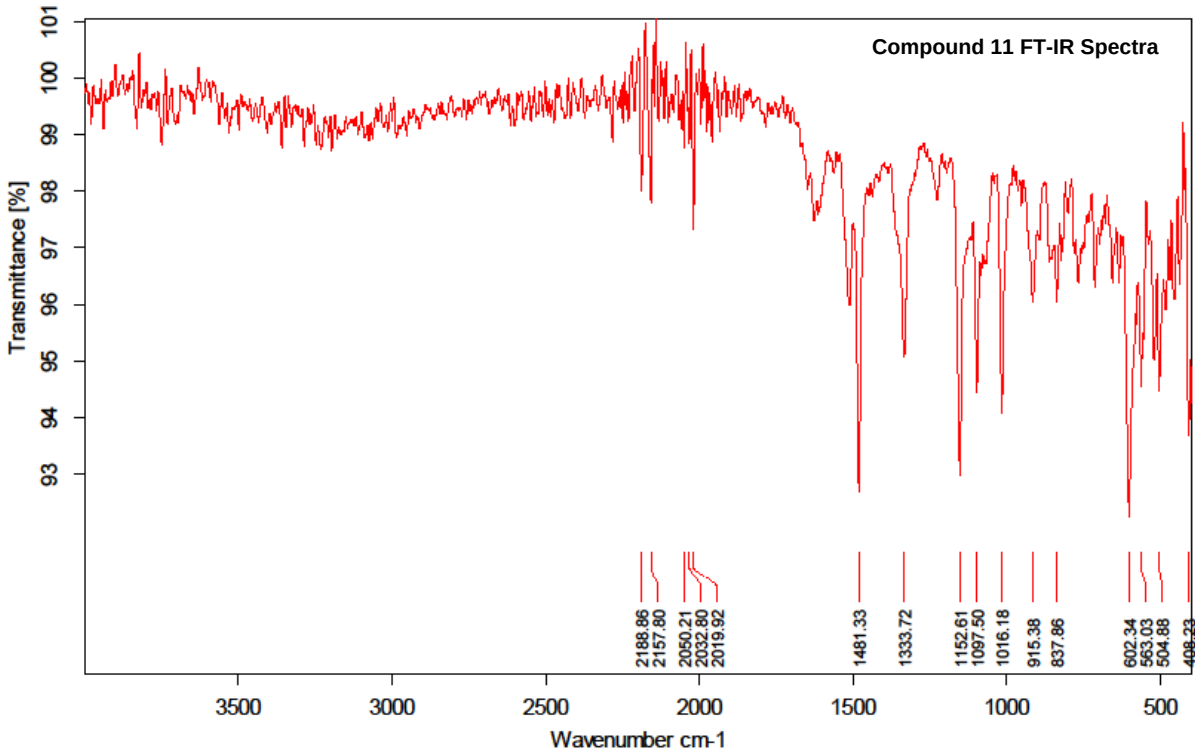

Compound 11 13C NMR Spectra
60000
55000
50000
45000
40000
35000
30000
25000
20000
15000
10000
5000
0
210 200 190 180 170 160 150 140 130 120 110 100 90 80 70 60 50 40 30 20 10 0 -10
f1 (ppm)

Compound 11 High-Res MS




VWD1 A, Wavelength=254 nm (TOBI 2019-10-19 13-11-04\052-0701.D)
mAU
800
600
400
200
0
23.762
20.529
0
5
10
15
20
25
30
35
min
Compound 11 Analytical
HPLC Trace (40 min)
Purity = 99.37%


VWD1 A, Wavelength=254 nm (TOBI 2019-10-19 13-11-04\052-0801.D)
mAU
1000
800
600
400
200
0
30.824
0
10
20
30
40
50
min
Compound 11 Analytical
HPLC Trace (60 min)
Purity = 99.37%

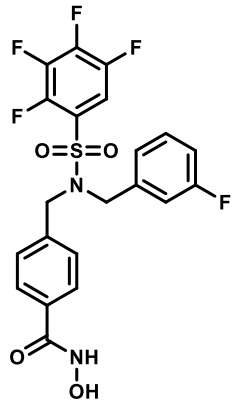


N-hydroxy-4-(((2,3,4,5-tetrafluoro-N-(3-fluorobenzyl)phenyl)sulfonamido)methyl)benzamide. Yield: 62%

Compound 12

$C_{21}H_{15}F_5N_2O_4S$

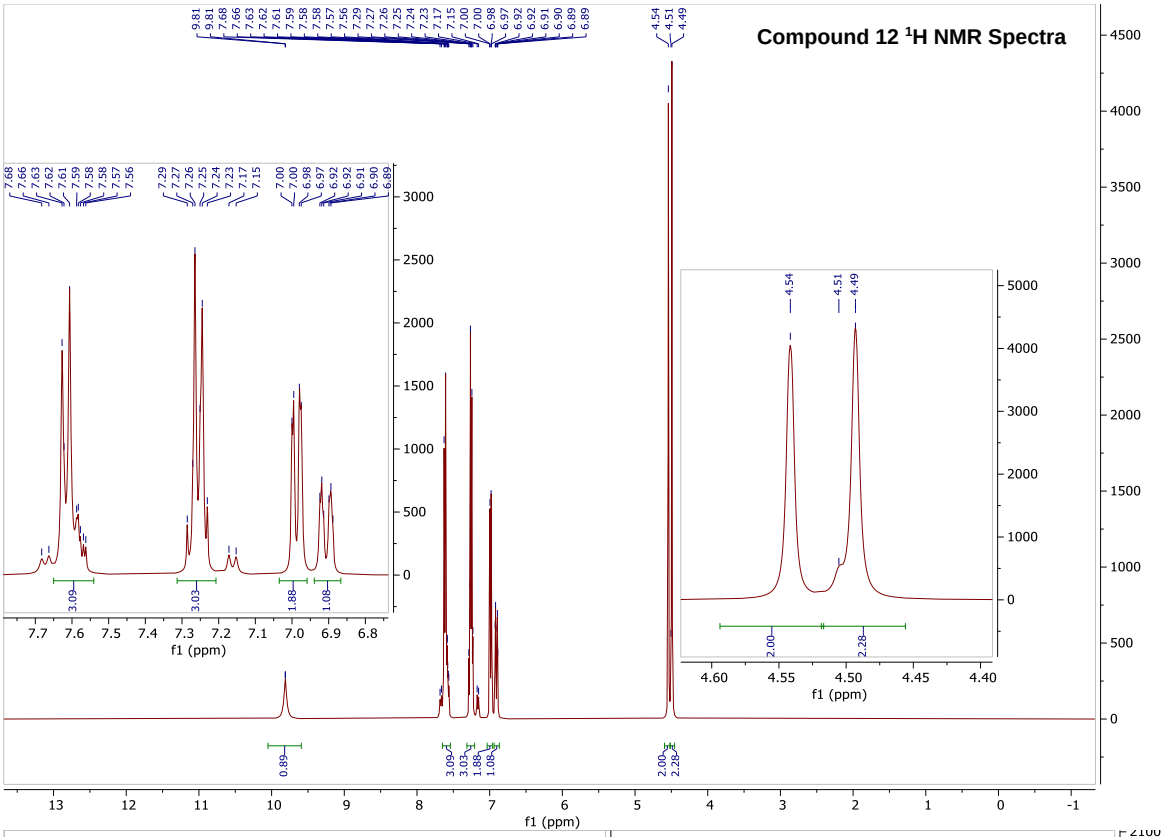
Mol. Weight: 486.41 $gmol^{-1}$

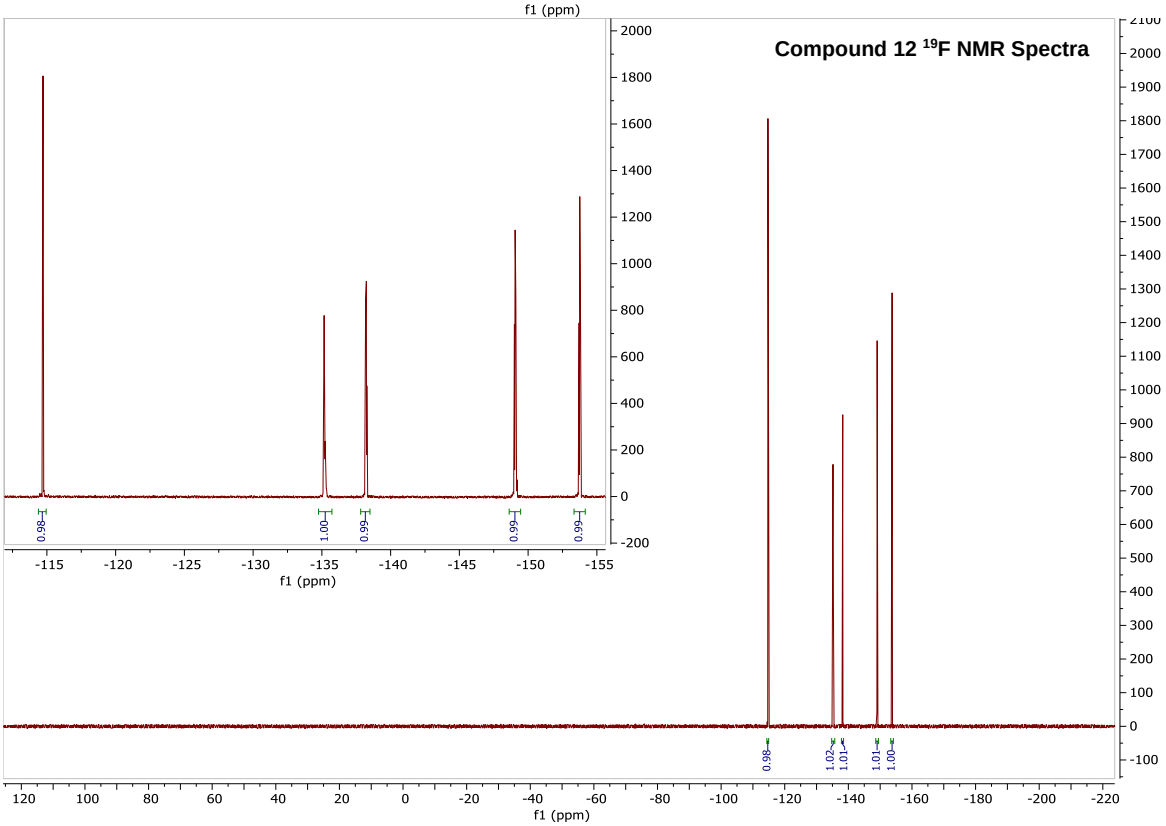


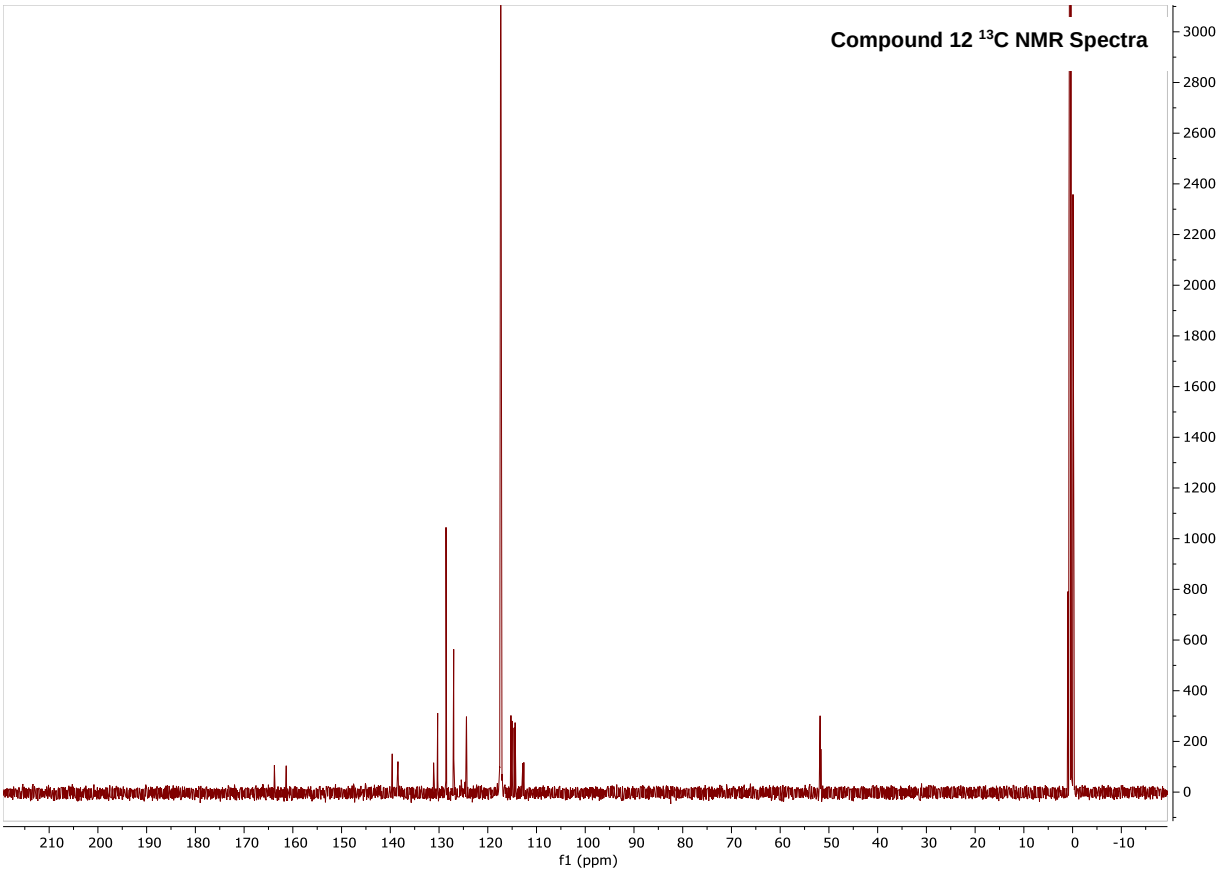
^{1}H NMR (400 MHz, CD_3CN) δ 10.05 – 9.59 (m, 1H), 7.65 – 7.54 (m, 3H), 7.31 – 7.21 (m, 3H), 6.99 (dd, *J* = 8.2, 2.0 Hz, 2H), 6.91 (dt, *J* = 10.1, 2.1 Hz, 1H), 4.54 (s, 2H), 4.49 (s, 2H).

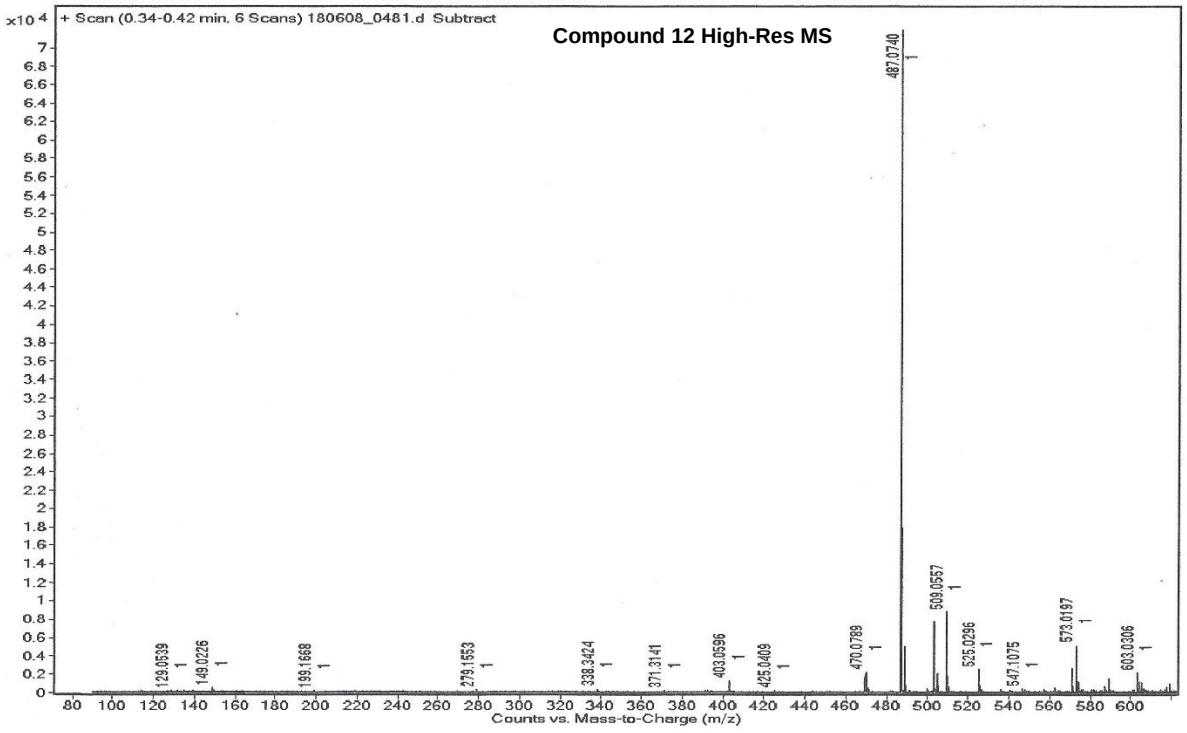
^{19}F NMR (376 MHz, CD_3CN) δ -114.4 – -115.0 (m), -135.2 (ddq, *J* = 21.0, 14.0, 7.2 Hz), -137.8 – -138.5 (m), -149.1 (ddd, *J* = 27.3, 18.7, 8.2 Hz), -153.3 – -154.2 (m).

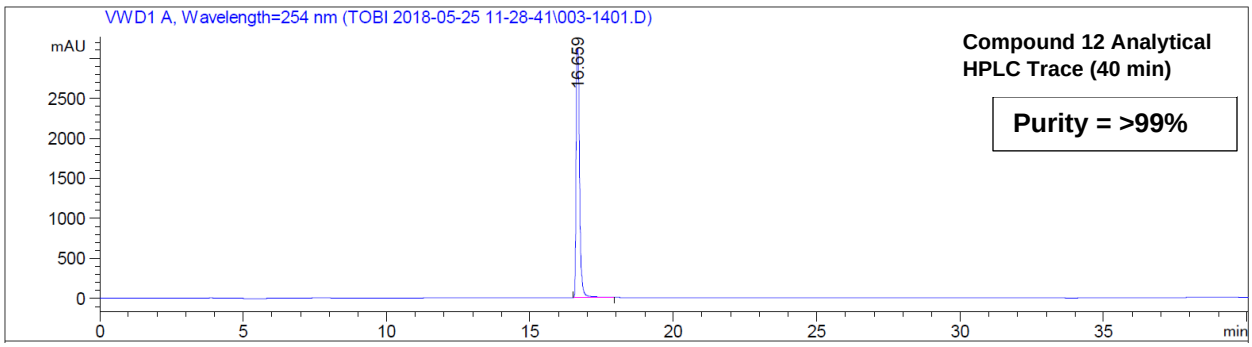
^{13}C NMR (101 MHz, CD_3CN) δ 163.8, 161.4, 139.6, 138.5 (d, *J* = 7.1 Hz), 131.1, 130.3 (d, *J* = 8.4 Hz), 128.6, 127.0, 124.4 (d, *J* = 2.8 Hz), 117.3, 115.1 (d, *J* = 22.0 Hz), 114.5 (d, *J* = 21.3 Hz), 112.7 (d, *J* = 22.0 Hz), 53.0 – 50.6 (m).

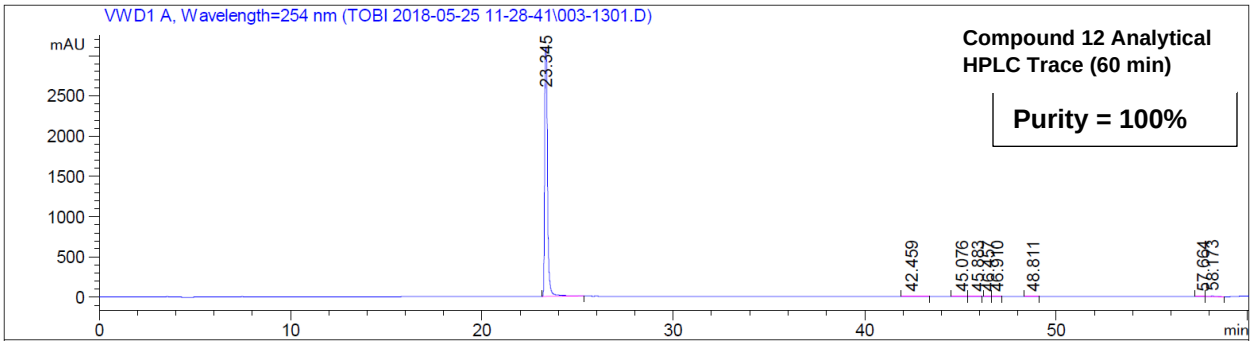

Compound 12 ^{1}H NMR Spectra
f1 (ppm)

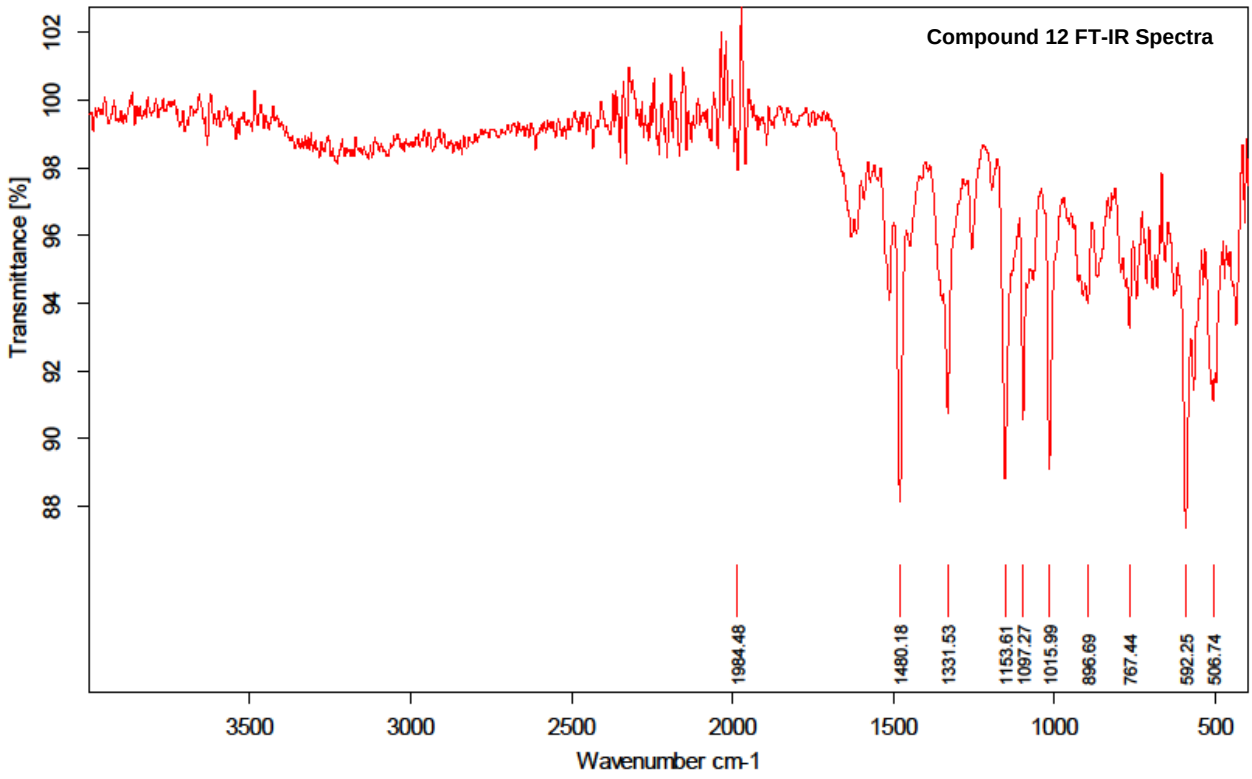

Compound 12 ^{19}F NMR Spectra
f1 (ppm)


Compound 12 ^{13}C NMR Spectra
f1 (ppm)


+ Scan (0.34-0.42 min, 6 Scans) 180608_0481.d Subtract
Compound 12 High-Res MS
Counts vs. Mass-to-Charge (m/z)


VWD1 A, Wavelength=254 nm (TOBI 2018-05-25 11-28-41\003-1401.D)
mAU
16.659
min
Compound 12 Analytical HPLC Trace (40 min)
Purity = >99%


VWD1 A, Wavelength=254 nm (TOBI 2018-05-25 11-28-41\003-1301.D)
mAU
23.345
42.459
45.076
48.811
57.664
58.173
min
Compound 12 Analytical HPLC Trace (60 min)
Purity = 100%

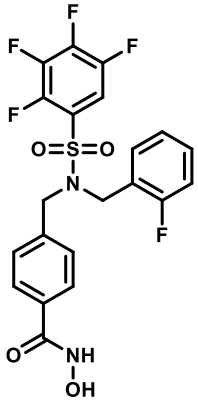

Compound 12 FT-IR Spectra
Transmittance [%]
Wavenumber cm-1
1984.48
1480.18
1331.53
1153.61
1097.27
1015.99
896.69
767.44
592.25
506.74

N-hydroxy-4-(((2,3,4,5-tetrafluoro-N-(2-fluorobenzyl)phenyl)sulfonamido)methyl)benzamide. Yield: 57%

Compound 13

$C_{21}H_{15}F_5N_2O_4S$

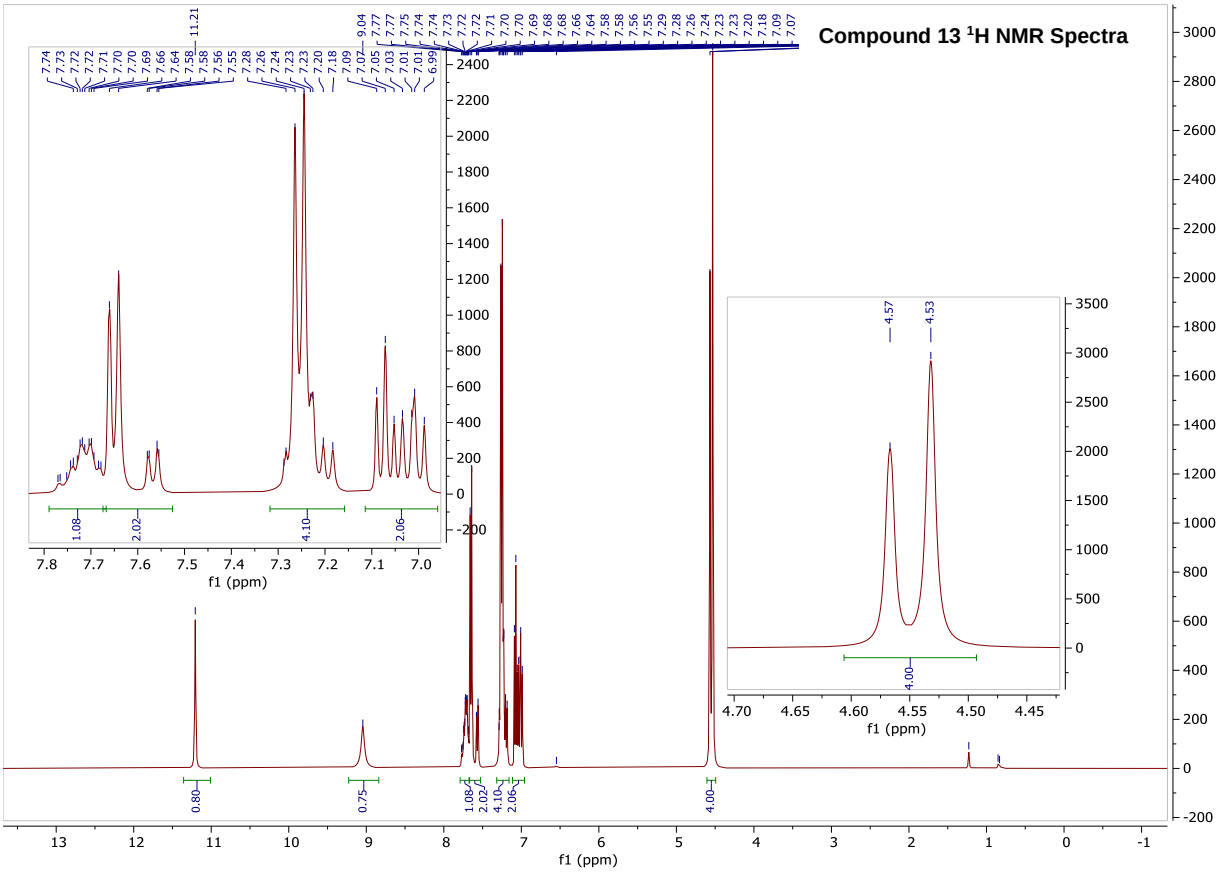
Mol. Weight: 486.41 $gmol^{-1}$

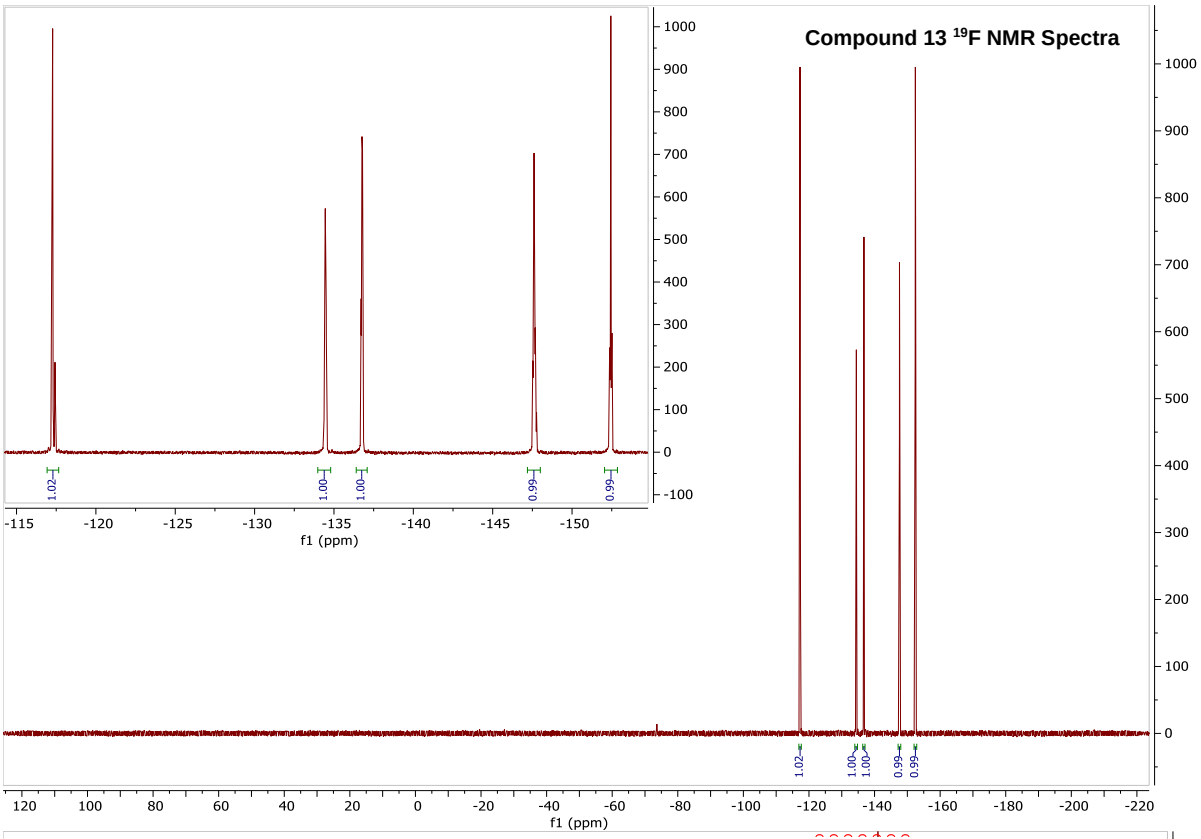


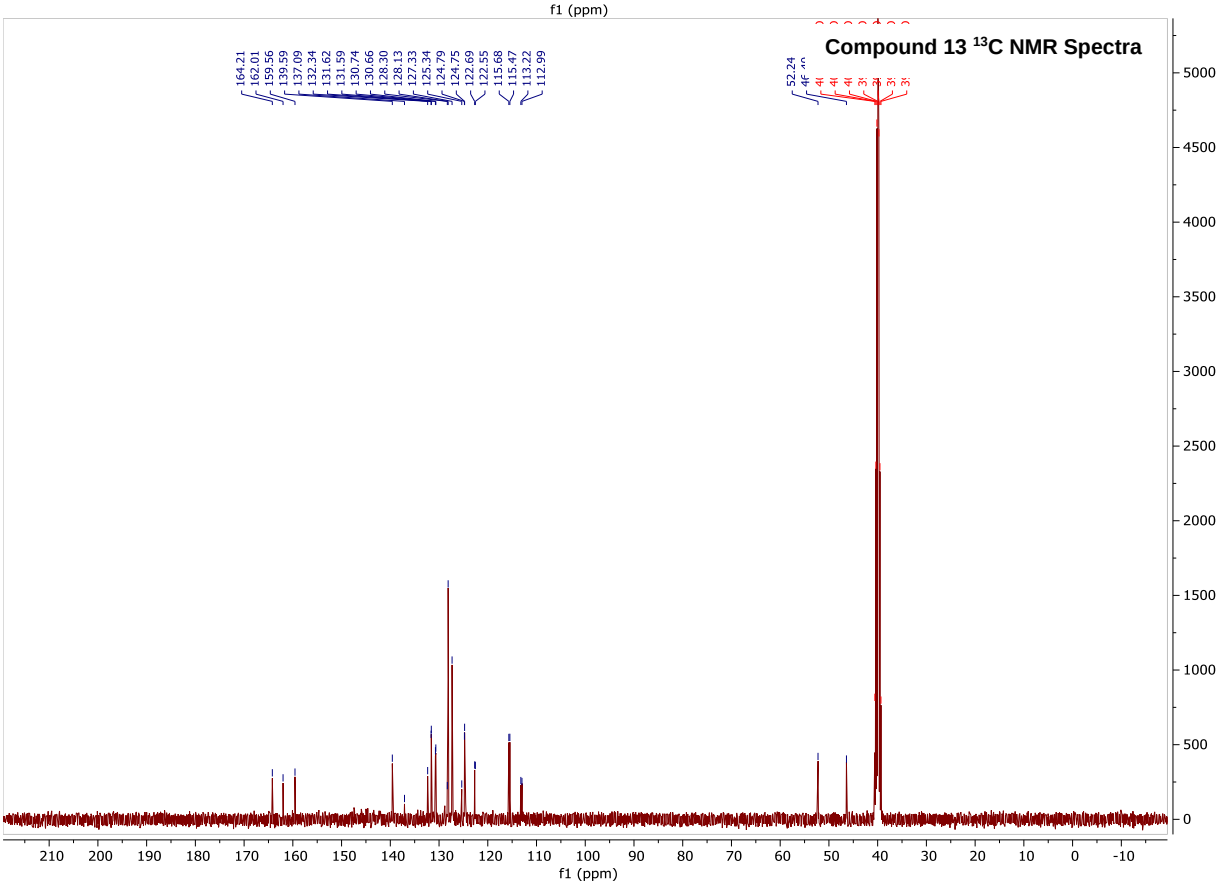
^{1}H NMR (400 MHz, DMSO) δ 11.21 (s, 1H), 9.04 (s, 1H), 7.72 (dddd, *J* = 17.8, 10.1, 4.9, 2.0 Hz, 1H), 7.67 – 7.53 (m, 2H), 7.32 – 7.16 (m, 4H), 7.11 – 6.96 (m, 2H), 4.55 (d, *J* = 13.9 Hz, 4H).

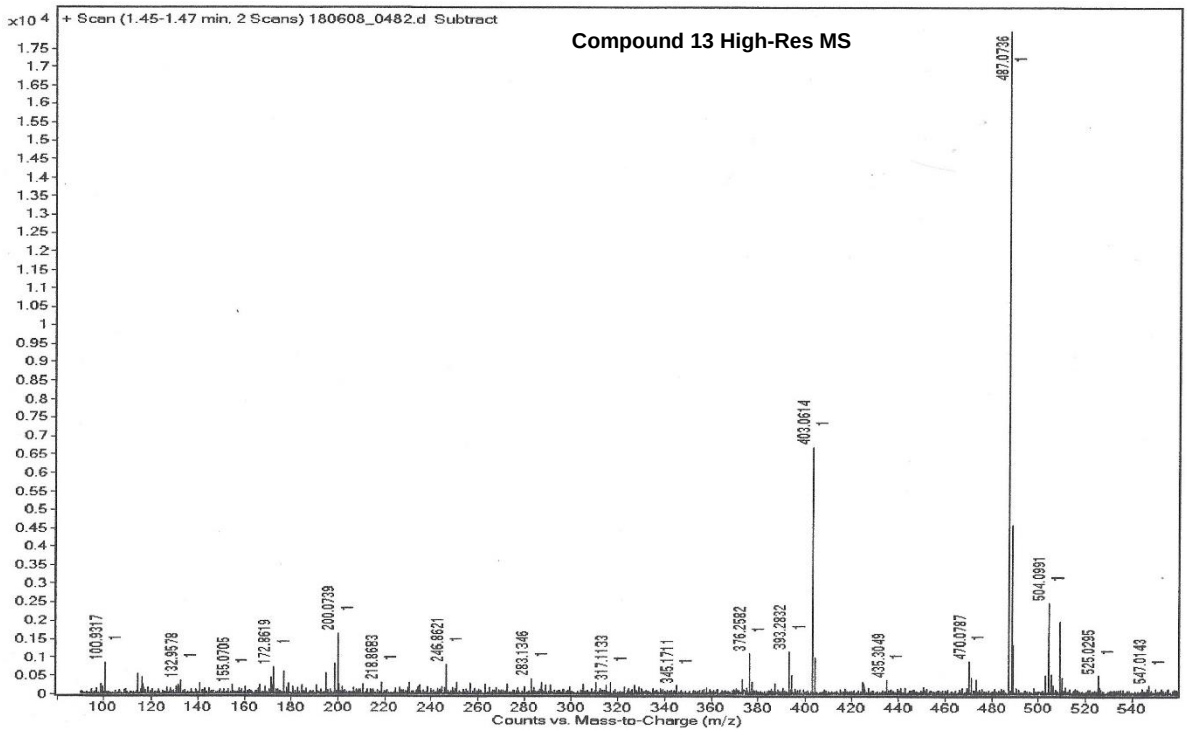
^{19}F NMR (376 MHz, DMSO) δ -117.3 (ddt, *J* = 58.6, 11.8, 6.5 Hz), -134.5 (ddt, *J* = 20.1, 13.2, 7.1 Hz), -136.8 (dt, *J* = 22.1, 11.2 Hz), -147.7 (dddd, *J* = 37.3, 29.9, 21.5, 8.2 Hz), -152.4 (t, *J* = 21.8 Hz).

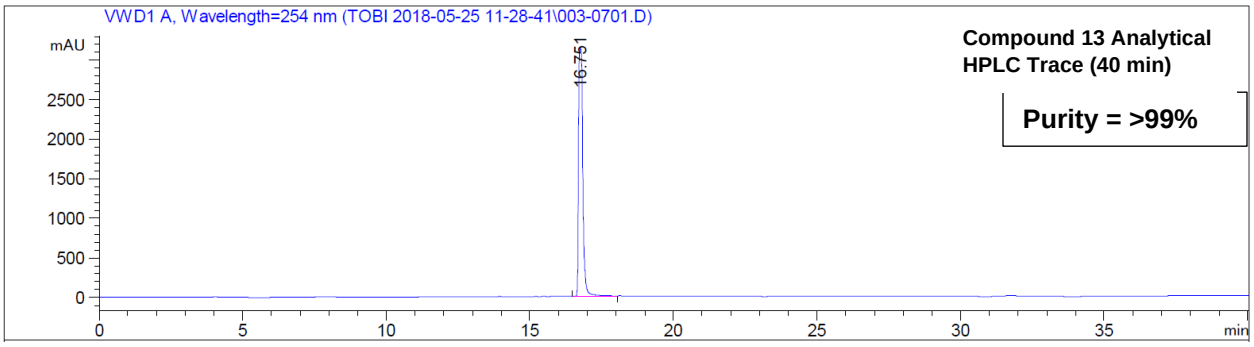
^{13}C NMR (101 MHz, DMSO) δ 164.2, 162.0, 159.7, 139.6, 137.1, 132.3, 131.6, 131.6, 130.7, 130.7, 128.3, 128.1, 127.3, 125.3, 124.8, 124.8, 122.7, 122.6, 115.7, 115.5, 113.2, 113.0, 52.2, 46.4.

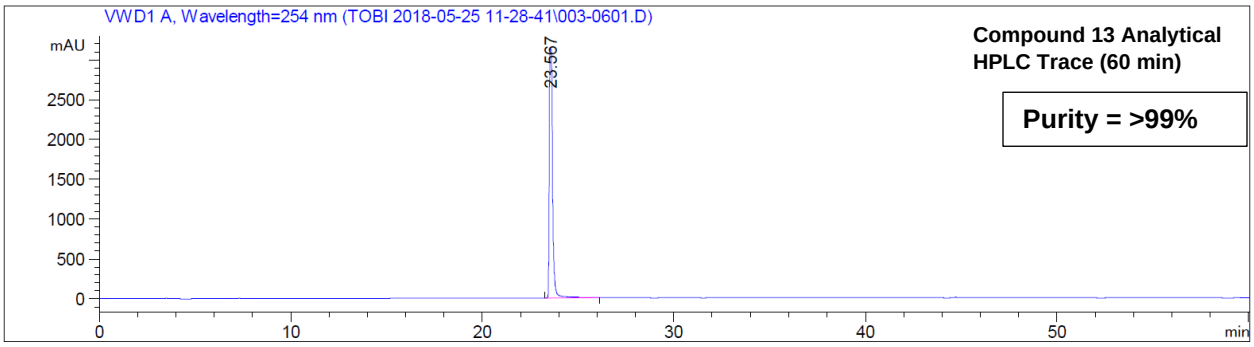


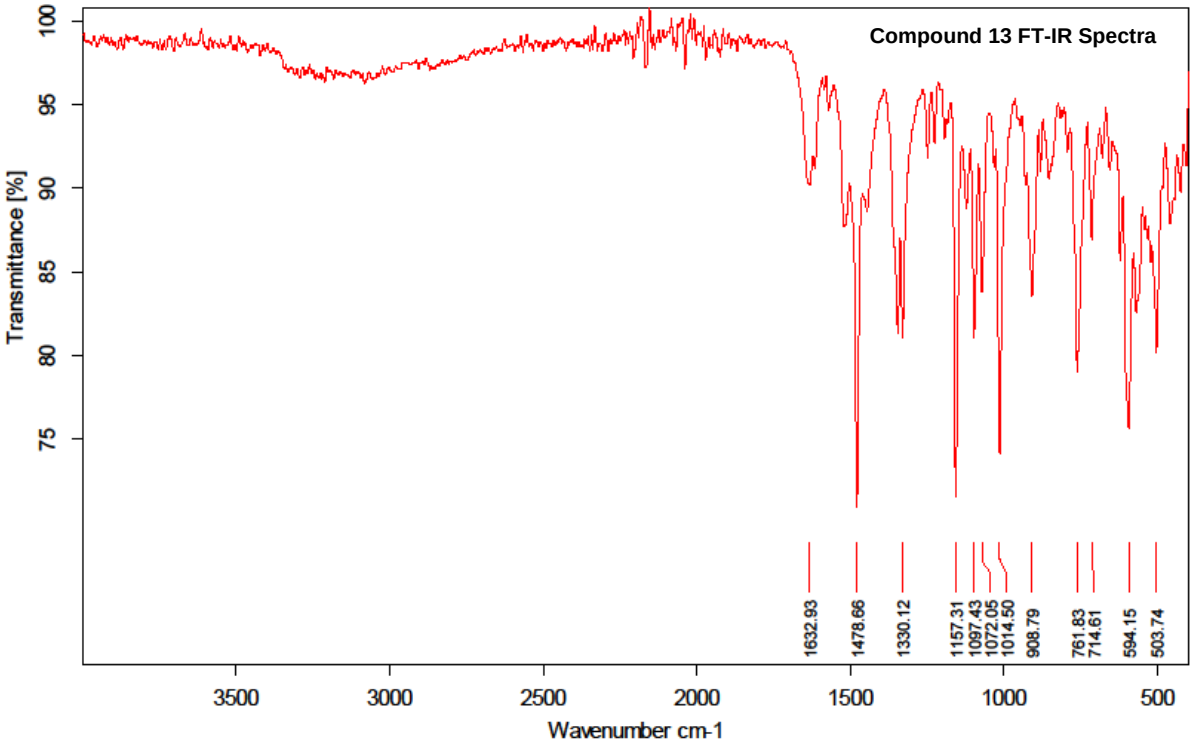

Compound 13 19F NMR Spectra
1.02
1.00
1.00
0.99
0.99
f1 (ppm)


Compound 13 13C NMR Spectra
164.21
162.01
159.56
139.59
137.09
132.34
131.62
131.59
130.74
130.66
128.30
128.13
127.33
125.34
124.79
124.75
122.69
122.55
115.68
115.47
113.22
112.99
52.24
f1 (ppm)


+ Scan (1.45-1.47 min, 2 Scans) 180608_0482.d Subtract
Compound 13 High-Res MS
x10 4
Counts vs. Mass-to-Charge (m/z)
100.9317
132.9578
155.0705
172.8619
200.0739
218.8683
246.8621
283.1346
317.1133
345.1711
376.2582
393.2832
403.0614
435.3049
470.0787
487.0736
504.0991
525.0295
547.0143


VWD1 A, Wavelength=254 nm (TOBI 2018-05-25 11-28-41\003-0701.D)
mAU
16.751
min
Compound 13 Analytical HPLC Trace (40 min)
Purity = >99%


VWD1 A, Wavelength=254 nm (TOBI 2018-05-25 11-28-41\003-0601.D)
mAU
23.567
min
Compound 13 Analytical HPLC Trace (60 min)
Purity = >99%

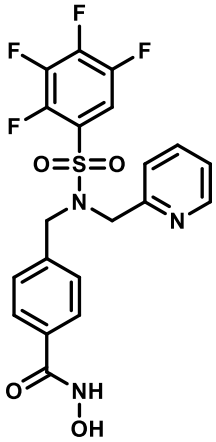


N-hydroxy-4-(((2,3,4,5-tetrafluoro-N-(pyridin-2-ylmethyl)phenyl)sulfonamido)methyl)benzamide.
Yield: 46%

Compound 14

$C_{20}H_{15}F_4N_3O_4S$

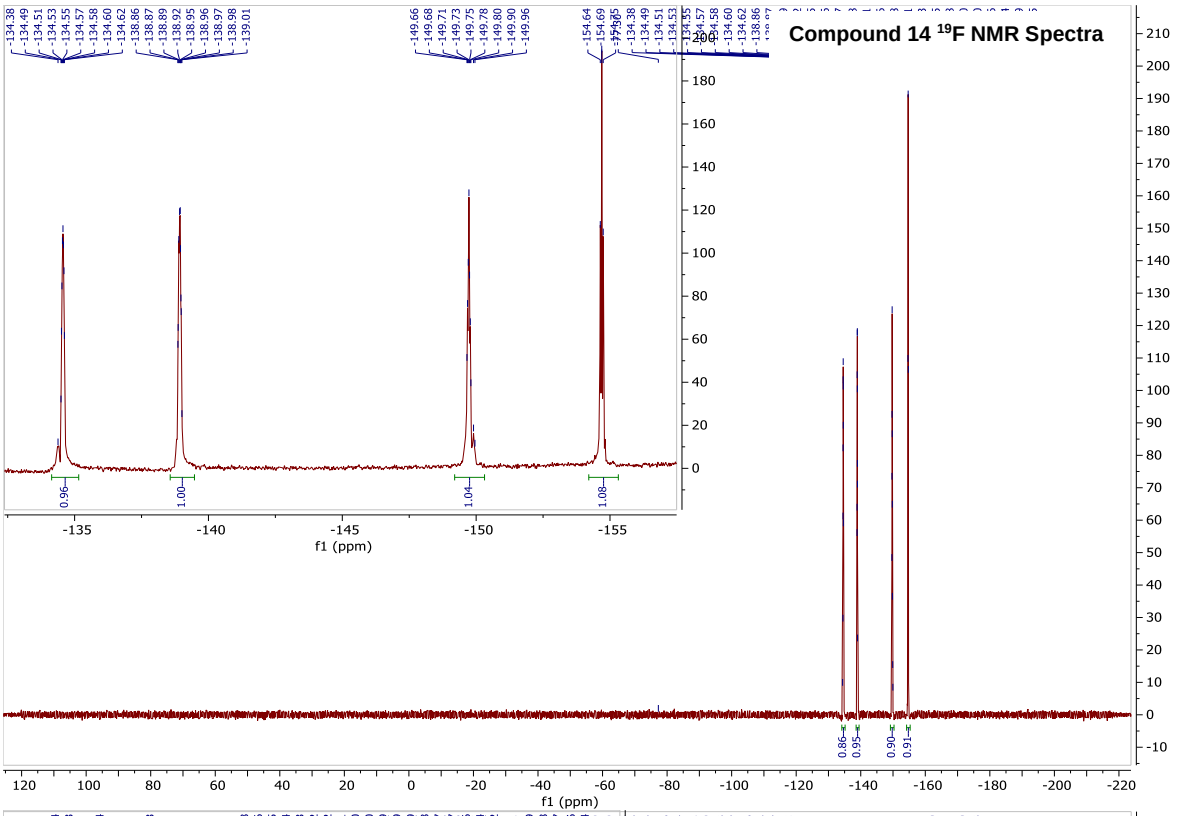
Mol. Weight: 469.41 $gmol^{-1}$

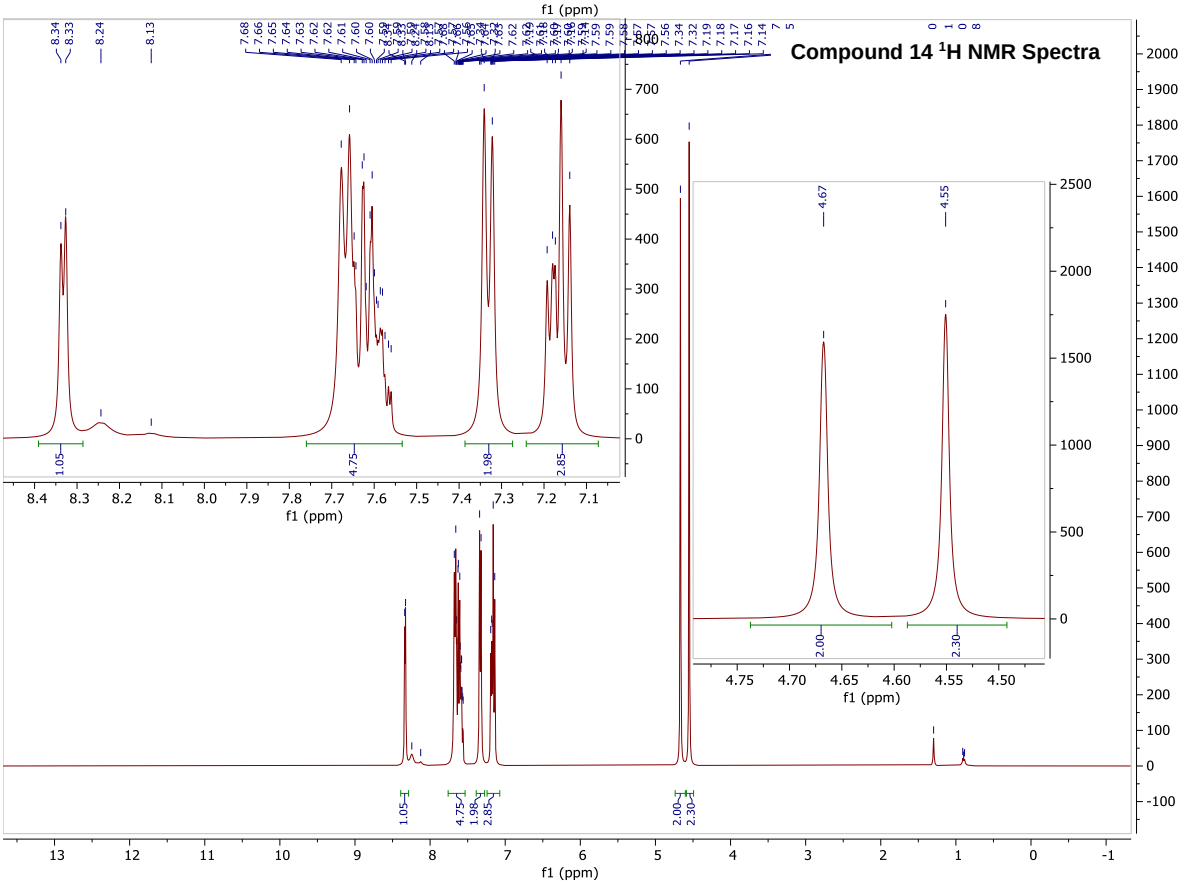


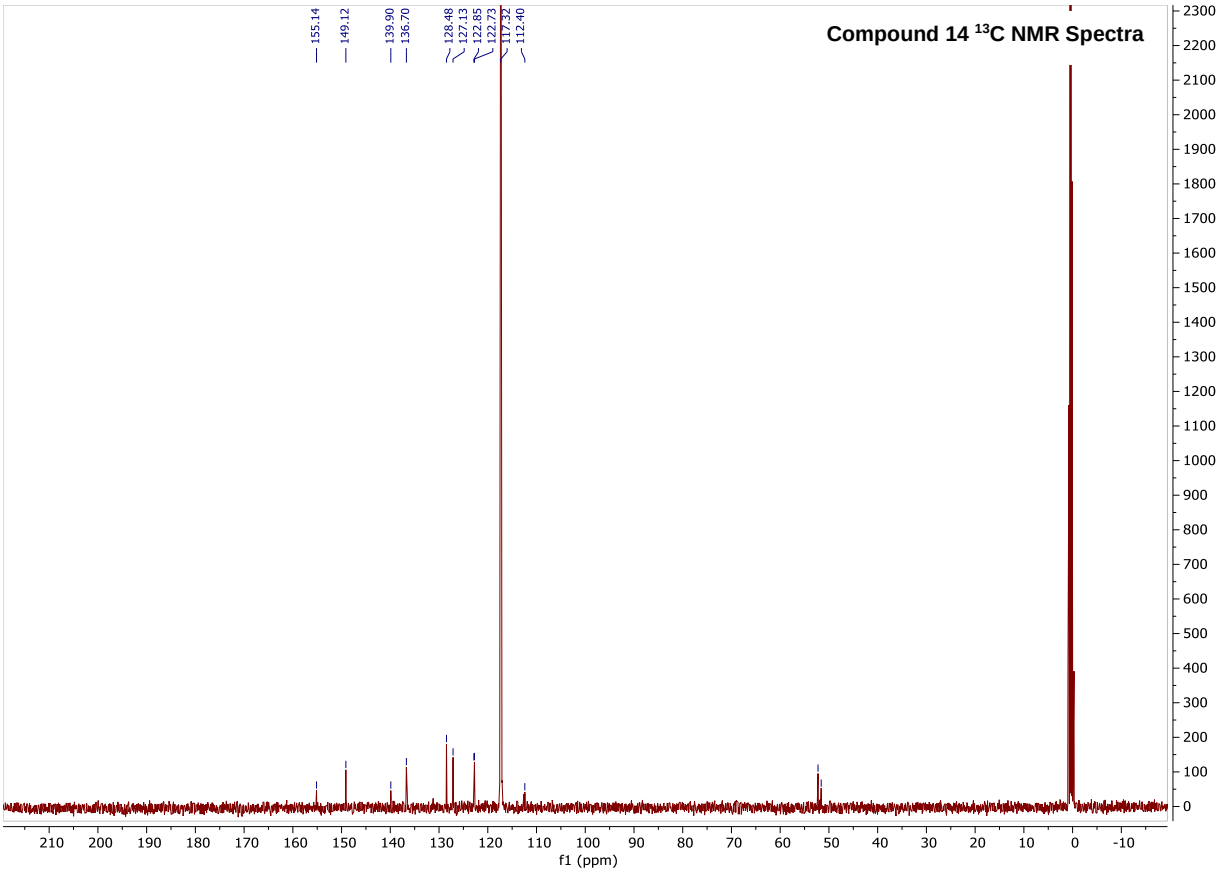
^{1}H NMR (400 MHz, CD_3CN) δ 8.33 (d, *J* = 4.6 Hz, 1H), 7.76 – 7.53 (m, 5H), 7.33 (d, *J* = 7.7 Hz, 2H), 7.24 – 7.07 (m, 3H), 4.67 (s, 2H), 4.55 (s, 2H).

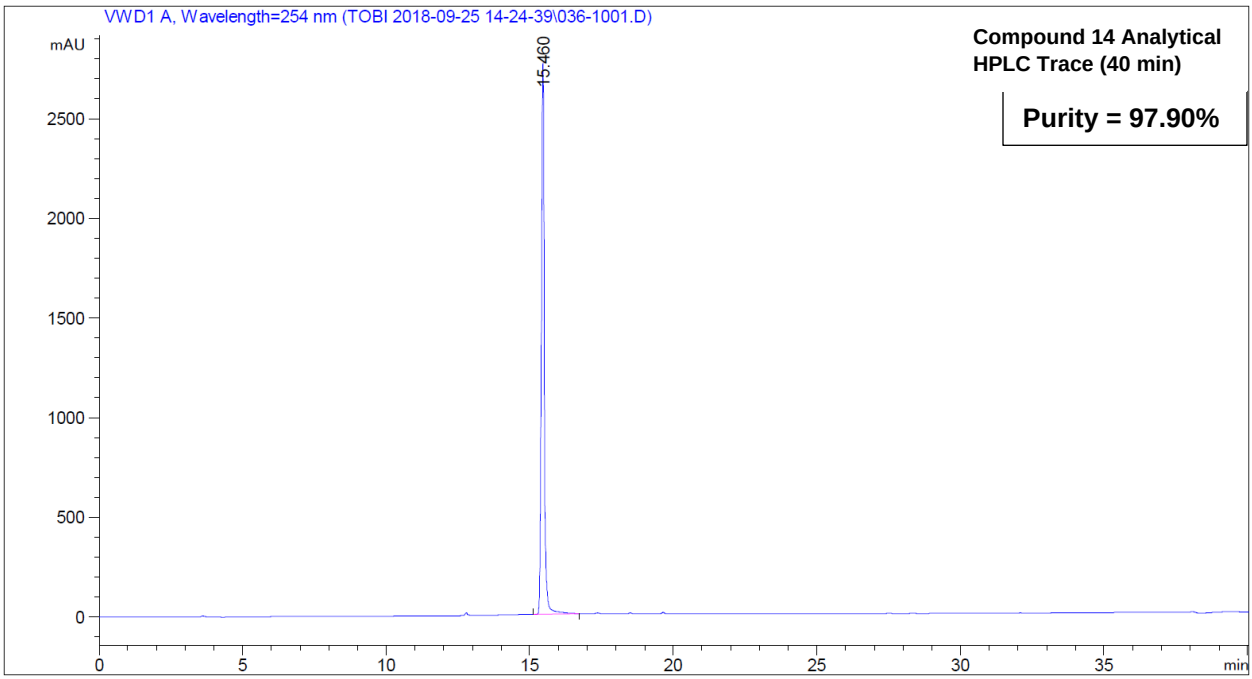
^{19}F NMR (376 MHz, CD_3CN) δ -134.6 (ddt, *J* = 20.6, 13.5, 6.9 Hz), -138.6 – -139.5 (m), -149.7 (ddd, *J* = 27.5, 18.8, 8.1 Hz), -154.7 (t, *J* = 20.5 Hz).

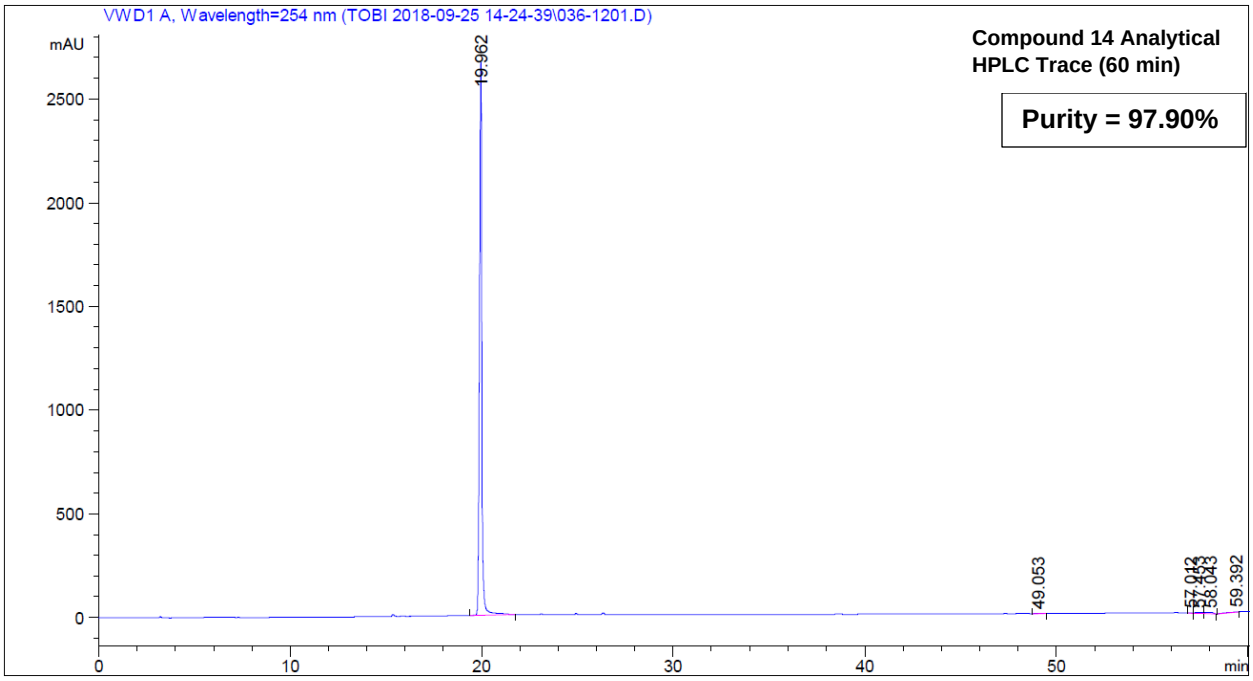
^{13}C NMR (101 MHz, CD_3CN) δ 155.1, 149.1, 139.9, 136.7, 128.5, 127.1, 122.8, 117.3, 112.4, 52.2, 51.6.

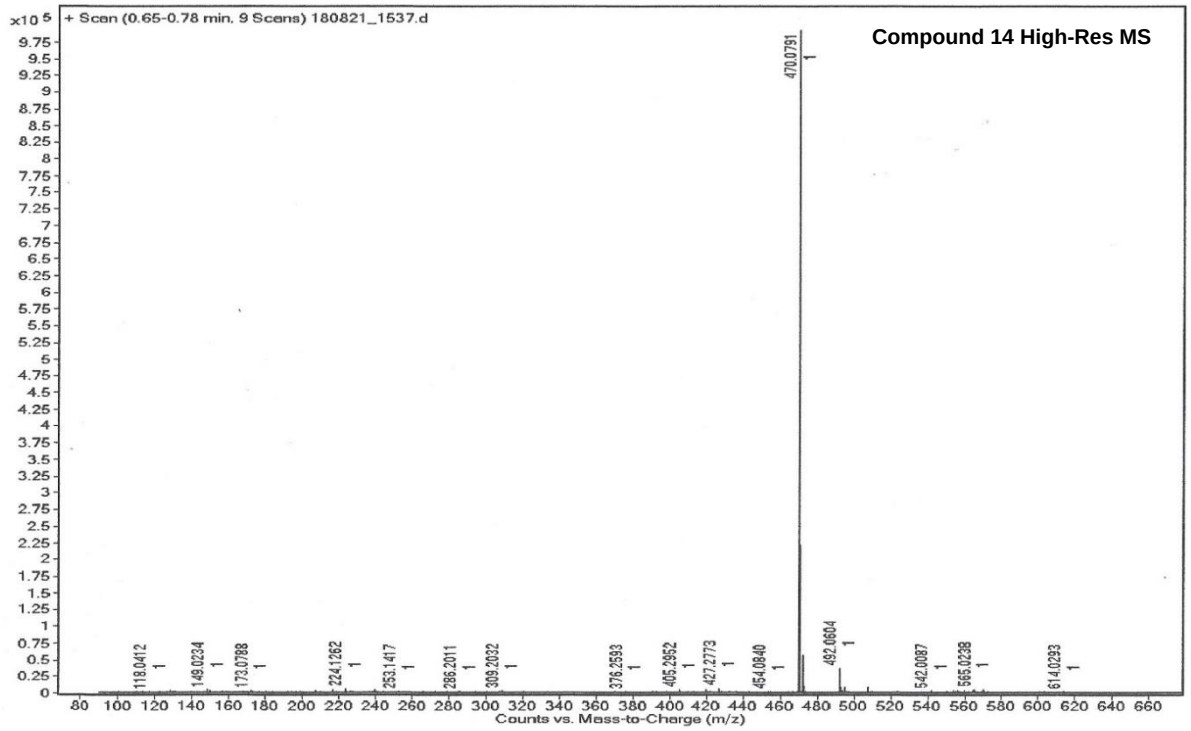

Compound 14 19F NMR Spectra
f1 (ppm)


Compound 14 1H NMR Spectra
f1 (ppm)


Compound 14 ^{13}C NMR Spectra
155.14
149.12
139.90
136.70
128.48
127.13
122.85
122.73
117.32
112.40
f1 (ppm)


VWD1 A, Wavelength=254 nm (TOBI 2018-09-25 14-24-39\036-1001.D)
Compound 14 Analytical HPLC Trace (40 min)
Purity = 97.90%
mAU
15.460
min


VWD1 A, Wavelength=254 nm (TOBI 2018-09-25 14-24-39\036-1201.D)
mAU
2500
2000
1500
1000
500
0
19.962
49.053
57.012
57.493
58.043
59.392
0
10
20
30
40
50
min
Compound 14 Analytical HPLC Trace (60 min)
Purity = 97.90%

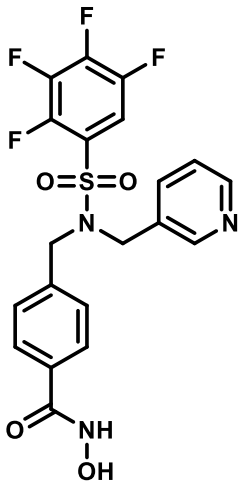

x10 5
+ Scan (0.65-0.78 min, 9 Scans) 180821_1537.d
470.0791
492.0604
118.0412
149.0234
173.0788
224.1262
253.1417
286.2011
309.2032
376.2593
405.2952
427.2773
454.0840
542.0087
565.0238
614.0233
Counts vs. Mass-to-Charge (m/z)
Compound 14 High-Res MS

N-hydroxy-4-(((2,3,4,5-tetrafluoro-N-(pyridin-3-ylmethyl)phenyl)sulfonamido)methyl)benzamide.
Yield: 43%

TO-317

$C_{20}H_{15}F_4N_3O_4S$

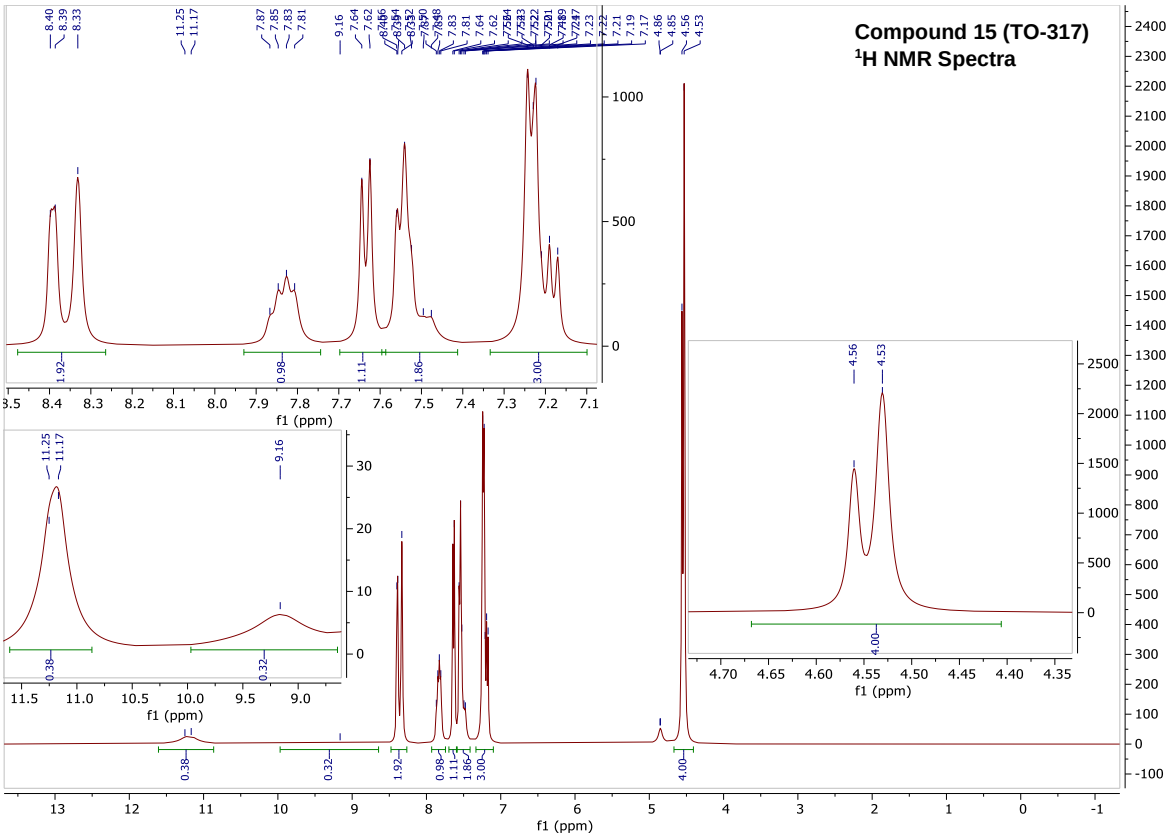
Mol. Weight: 469.41 $gmol^{-1}$

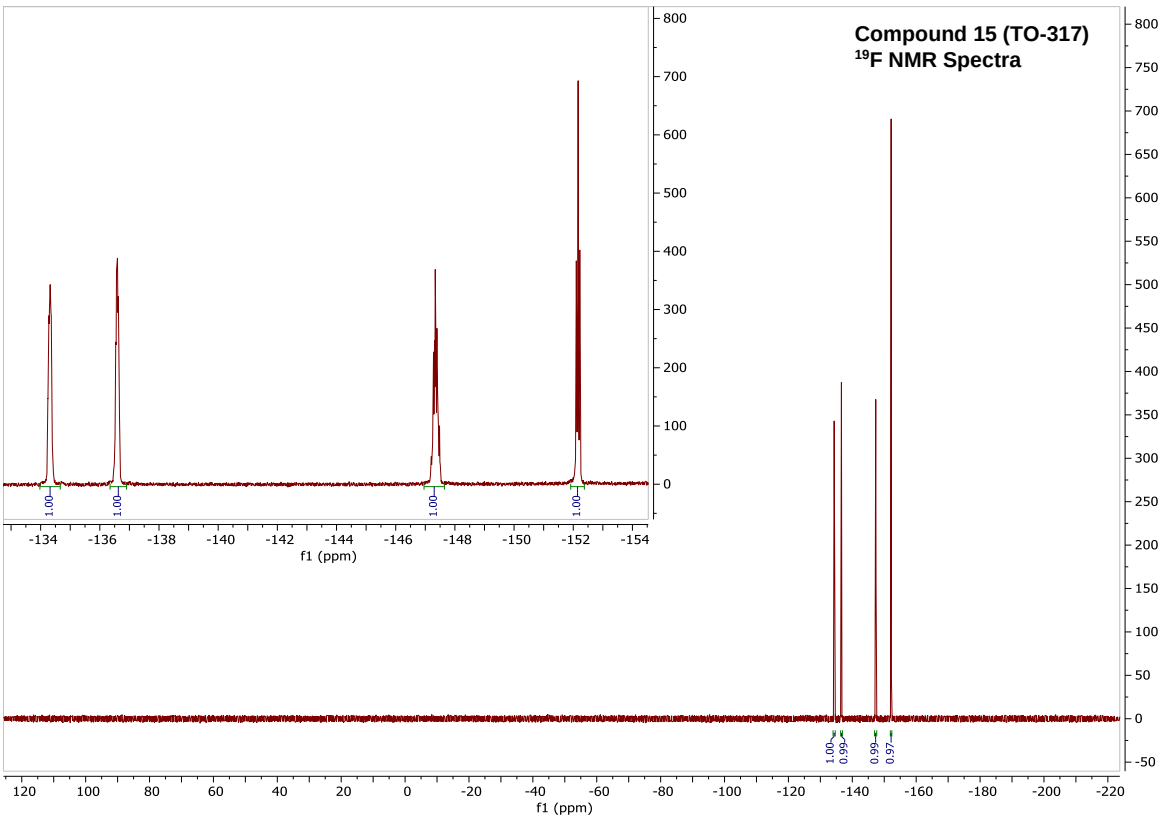


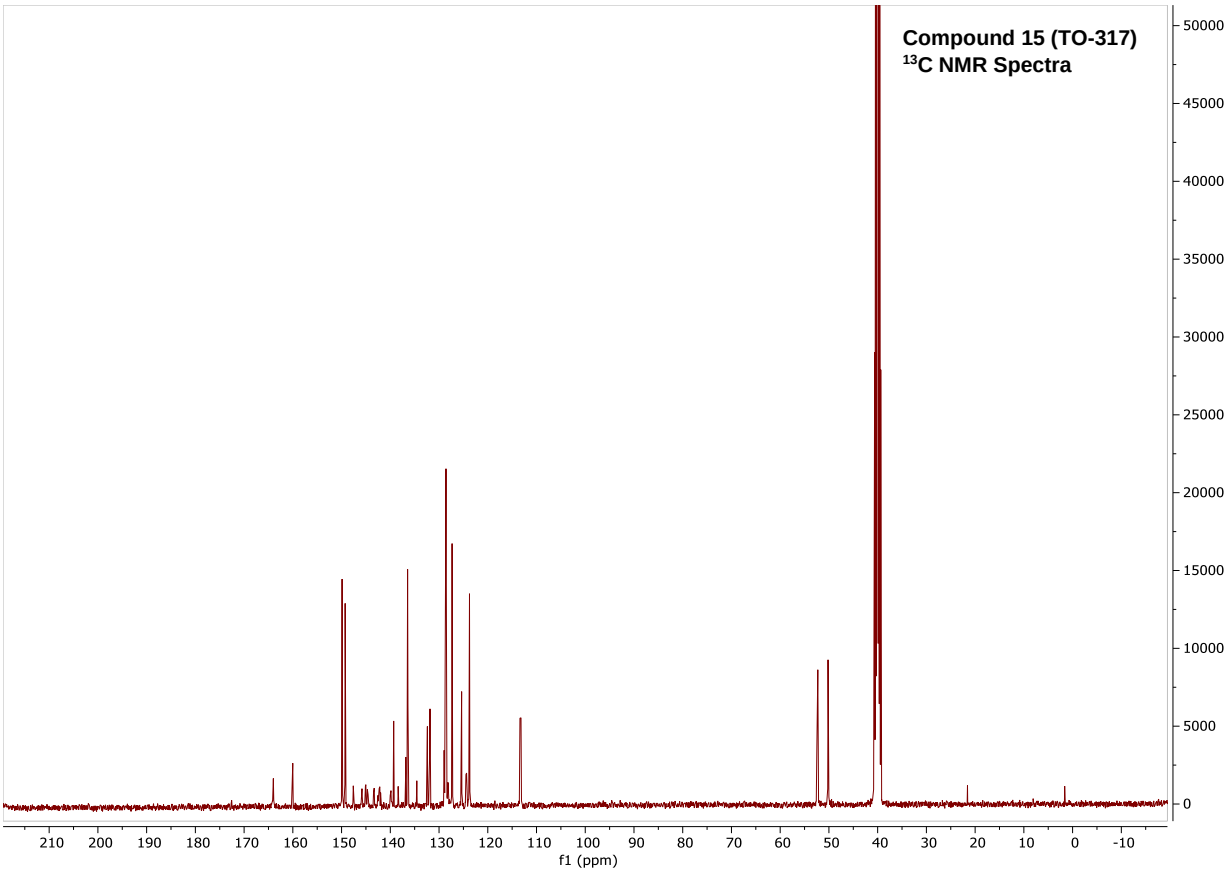
^{1}H NMR (400 MHz, DMSO) δ 11.21 (s, 1H), 9.16 (s, 1H), 8.48 – 8.26 (m, 2H), 7.93 – 7.74 (m, 1H), 7.63 (d, *J* = 7.9 Hz, 1H), 7.60 – 7.41 (m, 2H), 7.33 – 7.10 (m, 3H), 4.55 (d, *J* = 11.9 Hz, 4H).

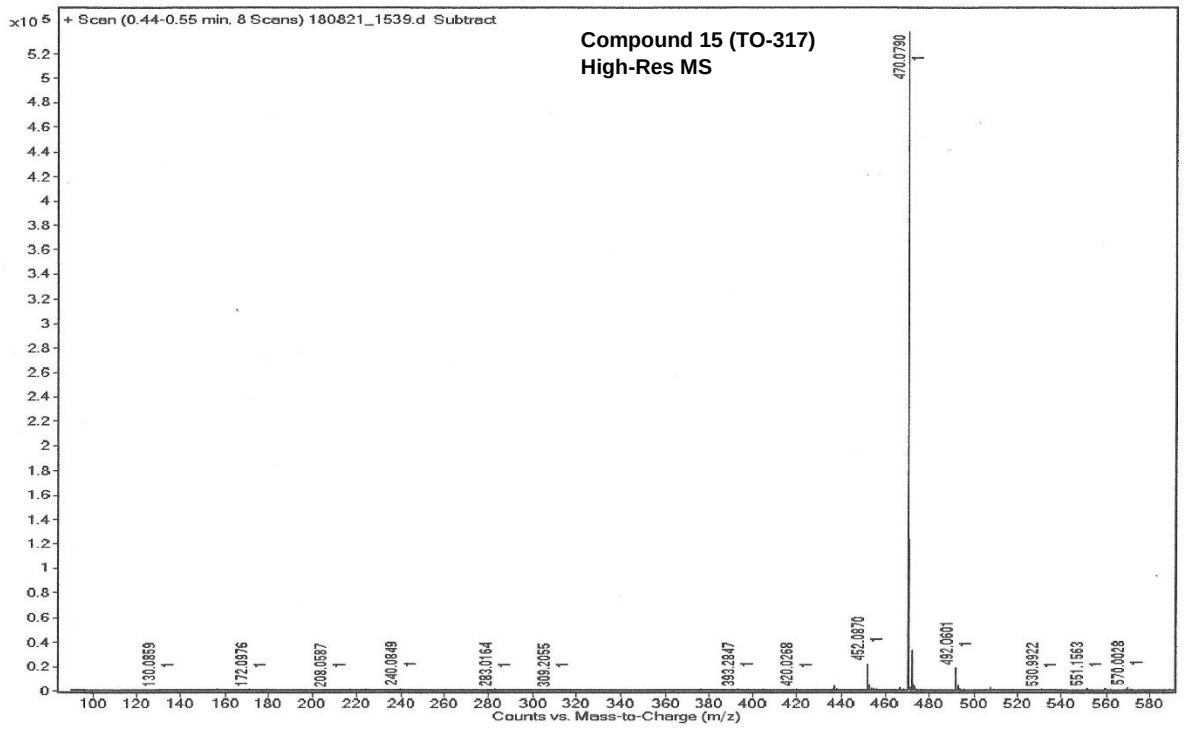
^{19}F NMR (376 MHz, DMSO) δ -134.3 (ddt, *J* = 20.7, 13.5, 7.2 Hz), -136.6 (dt, *J* = 21.8, 11.0 Hz), -147.4 (dtdd, *J* = 41.1, 30.5, 18.4, 9.7 Hz), -152.2 (t, *J* = 22.1 Hz).

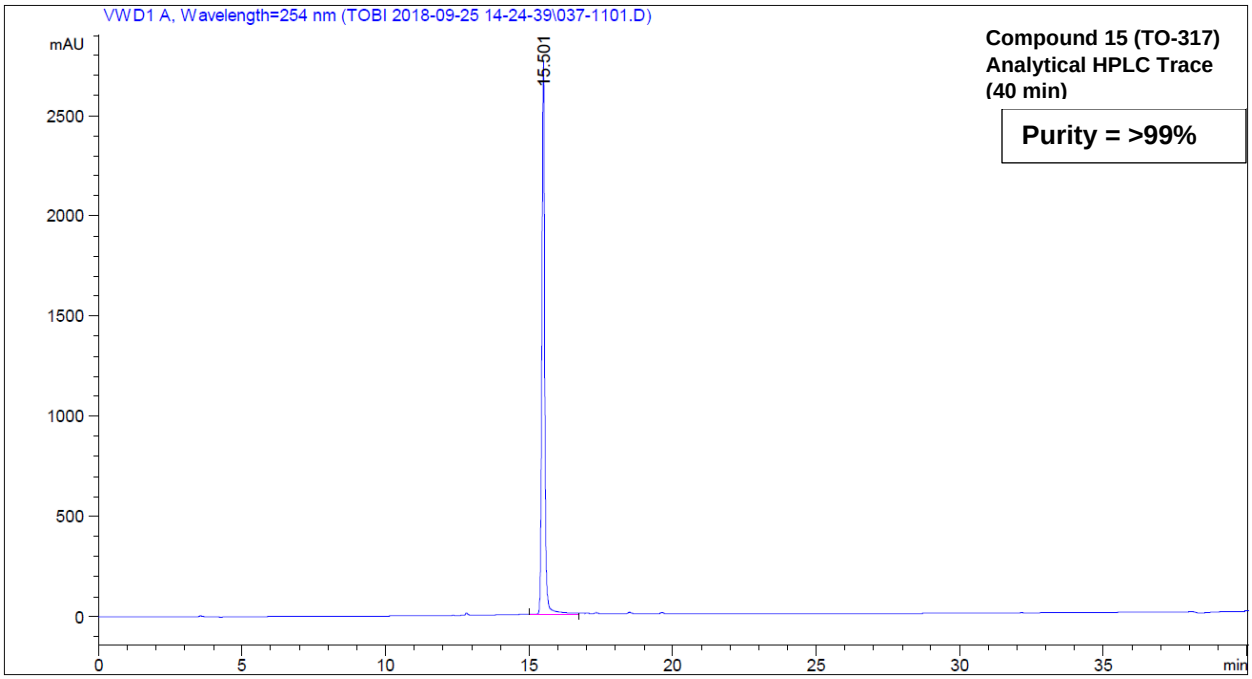
^{13}C NMR (101 MHz, DMSO) δ 164.0, 160.0, 149.9,147.6, 145.8, 144.6, 143.4, 142.4, 139.9, 138.4, 136.8, 134.56, 132.4, 132.0, 129.0, 127.3, 125.4, 124.5, 123.8, 113.4, 52.5, 50.2

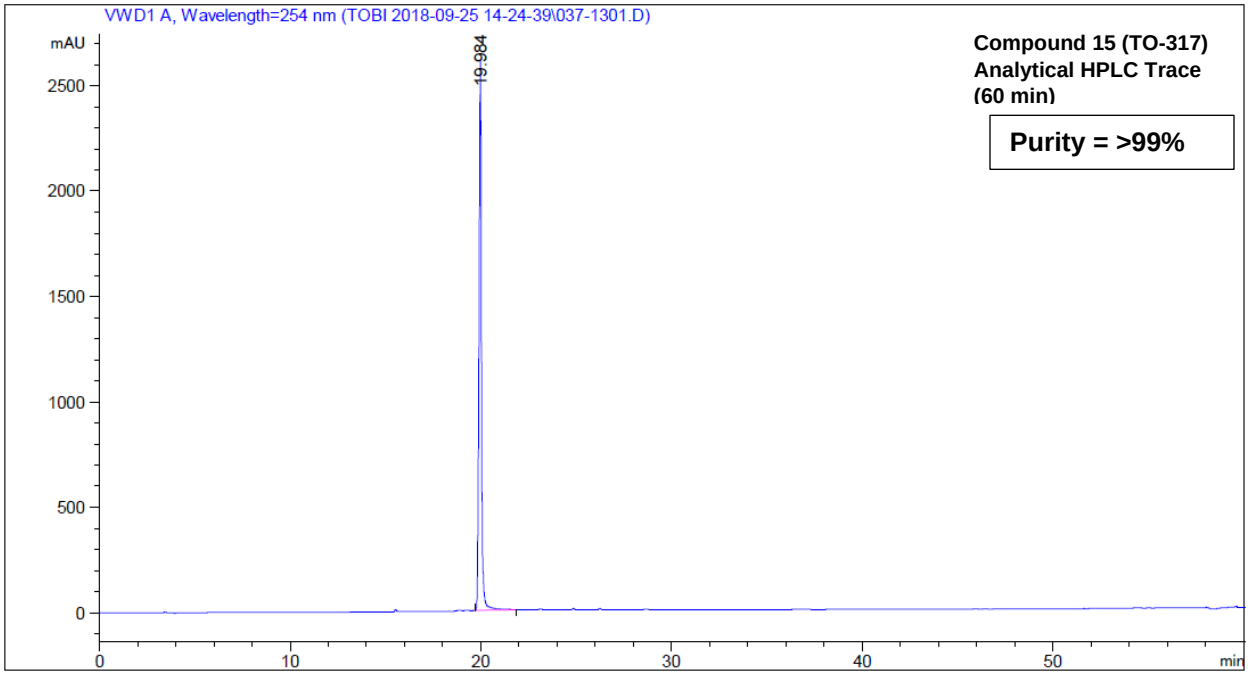

Compound 15 (TO-317)
^{1}H NMR Spectra
f1 (ppm)

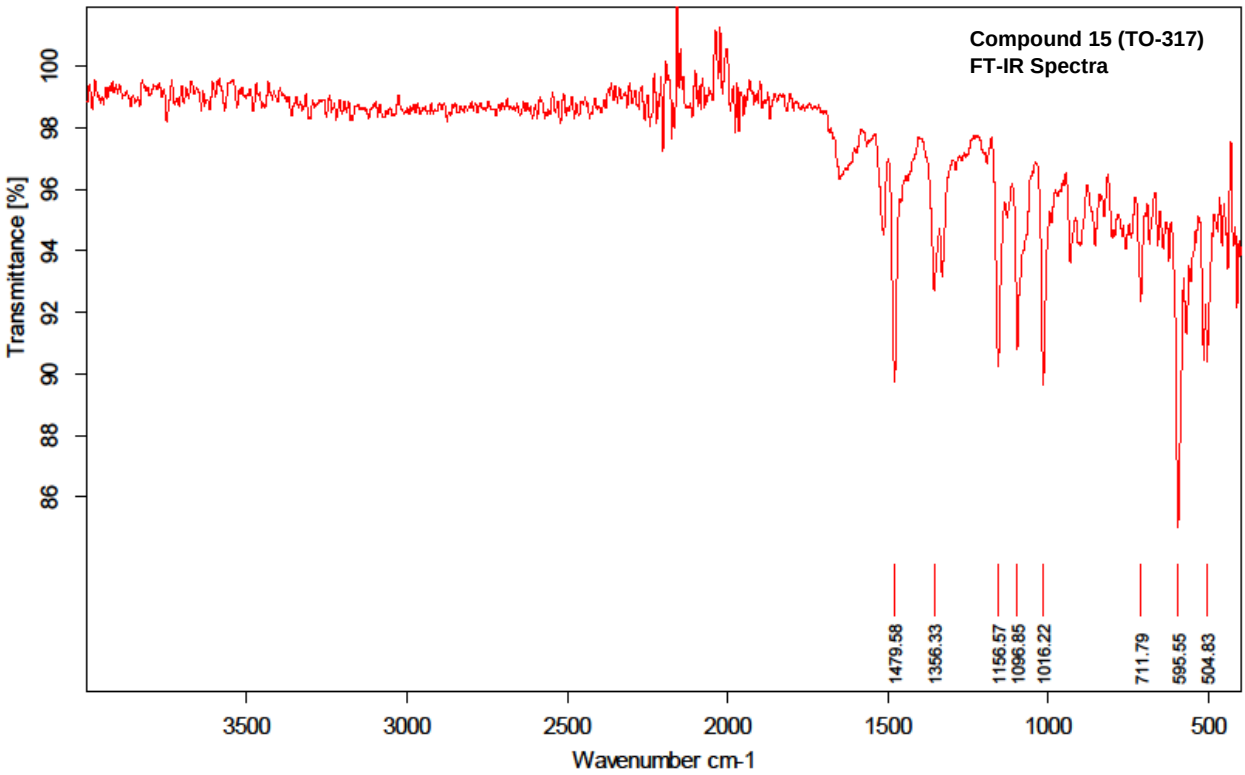

Compound 15 (TO-317)
^{19}F NMR Spectra
f1 (ppm)


Compound 15 (TO-317)
13C NMR Spectra
f1 (ppm)


+ Scan (0.44-0.55 min, 8 Scans) 180821_1539.d Subtract
Compound 15 (TO-317)
High-Res MS
Counts vs. Mass-to-Charge (m/z)


VWD1 A, Wavelength=254 nm (TOBI 2018-09-25 14-24-39\037-1101.D)
mAU
2500
2000
1500
1000
500
0
15.501
0
5
10
15
20
25
30
35
min
Compound 15 (TO-317)
Analytical HPLC Trace
(40 min)
Purity = >99%


VWD1 A, Wavelength=254 nm (TOBI 2018-09-25 14-24-39\037-1301.D)
mAU
2500
2000
1500
1000
500
0
19.984
0
10
20
30
40
50
min
Compound 15 (TO-317)
Analytical HPLC Trace
(60 min)
Purity = >99%


Compound 15 (TO-317)
FT-IR Spectra
Transmittance [%]
100
98
96
94
92
90
88
86
1479.58
1356.33
1156.57
1096.85
1016.22
711.79
595.55
504.83
3500
3000
2500
2000
1500
1000
500
Wavenumber cm-1

References

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