Supplementary Data

Discovery of OATD-01, a first-in-class, chitinase inhibitor as potential new therapeutics for idiopathic pulmonary fibrosis

Authors List:
Robert Koralewski,† Barbara Dymek,† Marzena Mazur,† Piotr Sklepkiewicz,† Sylwia Olejniczak,† Wojciech Czestkowski,† Krzysztof Matyszewski,† Gleb Andryianau,† Piotr Niedziejko,† Michal Kowalski,† Mariusz Gruza,† Bartłomiej Borek,† Karol Jedrzejczak,† Agnieszka Bartoszewicz,† Elżbieta Pluta,† Aleksandra Rymaszewska,† Magdalena Kania,† Tomasz Rejczak,† Sylwia Piasecka,† Michal Mlacki,† Marcin Mazurkiewicz,† Michał Piotrowicz,†, ‖ Magdalena Salamon,† Agnieszka Zagozdzon,† Agnieszka Napiorkowska-Gromadzka,§ Aneta Bartlomiejczak,§ Witold Mozga,† Paweł Dobrzański,† Karolina Dzwonek,† Jakub Golab,†, ‡ Marcin Nowotny,§ Jacek Olczak,† Adam Golebiowski*,†

†OncoArendi Therapeutics SA, Żwirki i Wigury 101, 02-089 Warsaw, Poland
‡Department of Immunology, Medical University of Warsaw, Nielubowicza 5, 02-097 Warsaw, Poland
§Structural Biology Center, International Institute of Molecular and Cell Biology, Trojdena 4, 02-109 Warsaw, Poland

Intestinal permeability results of OATD-01 using Caco-2 cell line.

Mean Papp (10-6 cm/s)		Mean efflux ratio	Mean recovery %	
A to B	**B to A**		**A to B**	**B to A**
0.58	17.6	30.2	57.0	75.9

Papp: apparent permeability coefficient.

Physicochemical properties of OATD-01

$logD_{7.4}$[a]	2.4
PSA[b]	83
pKa[c]	3.50 ± 0.01 and 6.48 ± 0.02
Solubility mg/mL	0.08 in water at pH 7
	42.9 in citrate buffer at pH 1

[a] Distribution coefficient between 1-octanol and aqueous phosphate buffer measured at pH 7.4

[b] PSA (polar surface area) was calculated by ChemDraw

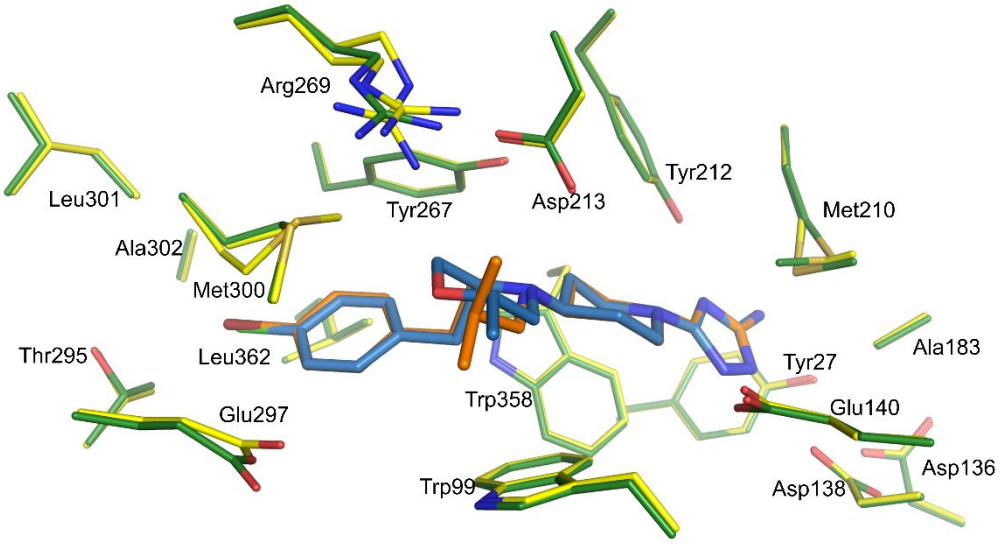
[c] pKa was determined from the potentiometric data collected, by Yasuda-Shedlovsky extrapolation

No significant inhibition of any CYP tested

Cytochrome	IC_{50} [µM]
CYP1A2	>50
CYP2C9	>50
CYP2C19	>50
CYP2D6	>50
CYP3A4	>50
CYP2B6	>50
CYP2C8	>50

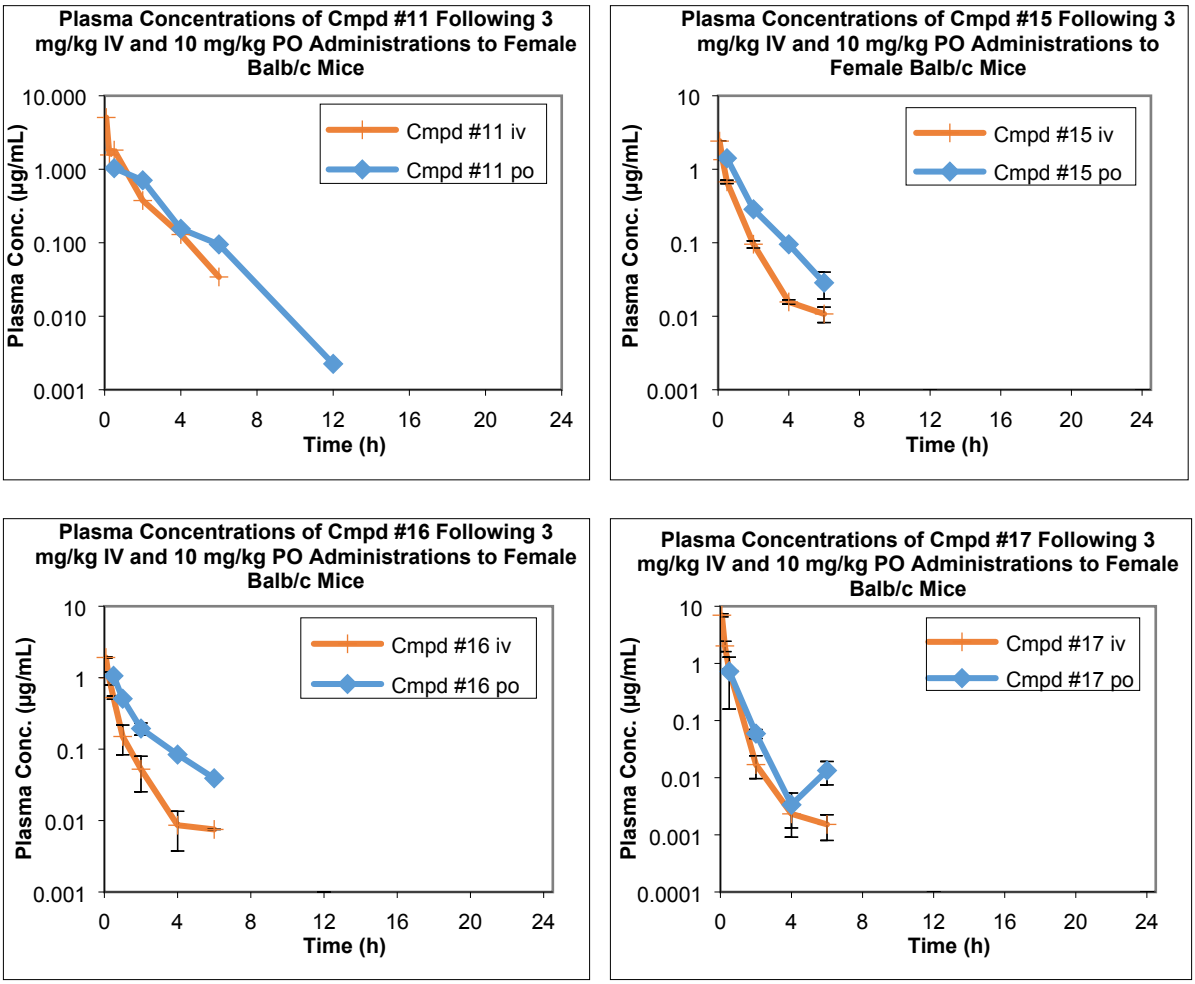
No significant inhibition of any transporter tested

Transporter	% of inhibition	
	At 1µM	At 10 µM
OCT2	-5.2	-0.6
ASBT	-24.4	-14.6
BCRP	1.6	1.5
MRP1	-3.0	-0.9
MRP2	-1.9	1.2
MRP3	0.2	0.0
NTCP	-8.2	5.5
OAT1	-16.9	-31.0
OAT3	-5.3	3.2
OATP1B1	-25.5	0.9
OCT1	-2.8	18.5
OATP1B3	-23.5	-15.1
P-gp	-0.7	0.7



Sl Figure 1. Superimposition of the active site residues in hCHIT1 complexed to **OATD-01** (PDB code 6ZE8, ligand carbons colored in blue and hCHIT1 residues shown as green sticks) and **OAT-177** (PDB code 5NRA, ligand carbons colored in orange and hCHIT1 residues shown as yellow sticks). The binding mode is largely the same for both compounds. The key difference is stabilization of Arg269 and Met300 sidechains by the interaction with 2-methylmorpholine ring of **OATD-01**.

PK profiles of compounds 11, 15-17



SI Figure 2. Plasma concentration time-course (log-linear) for compounds **11**, **15**, **16**, and **17** after single intravenous 3 mg/kg (iv) bolus and after 10 mg/kg oral administration (po) in female BALB/c mice.

SI Table 1. Mean pharmacokinetic parameters of compounds **11**, **15**, **16**, and **17** in mice (Balb/c; female) determined by the non-compartmental model (NCA).

PK parameter	Cmpd #							
	11		**15**		**16**		**17**	
Route	iv	po	iv	po	iv	po	iv	po
Dose (mg/kg)	3	10	3	10	3	10	3	10
$AUC_{0\text{-}24}$ (mg*h/L)	3.88	2.97	1.52	2.13	0.97	1.41	2.52	0.85
$AUC_{0\text{-}24}$/D (kg*mg*h/L*mg)	1.29	0.297	0.51	0.21	0.32	0.14	0.84	0.08
C_0 or C_{max} (mg/L)	9.08	1.03	3.23	1.41	2.66	1.06	12.91	0.72
C_{max}/D (kg*mg/L*mL)	n/a	0.10	n/a	0.14	n/a	0.106	n/a	0.072
T_{max} (h)	n/a	0.5	n/a	0.5	n/a	0.5	n/a	0.5
$T_{½}$ (h)	1.16	1.26	0.92	1.20	1.13	1.73	1.15	0.89
CL (L/h/kg)	0.77	n/a	1.98	n/a	3.09	n/a	1.19	n/a
Vss (L/kg)	0.78	n/a	1.33	n/a	2.06	n/a	0.29	n/a
Bioavailability (%)	n/a	22.93	n/a	42.19	n/a	43.74	n/a	10.06
formulation	10% EtOH/10% solutol/80% water							

Increasing the size of the 2-substituent (e.g. compounds **11-17**) led in general to equipotent inhibitors, but with much inferior PK (e.g. compounds **11, 15, 16, 17 –** SI Fig. 2 and SI Table 1). In contrast to **OATD-01**, compounds **11**, **15**, **16**, and **17** showed significantly higher clearance values after single iv 3 mg/kg dose in Balb/c mice. Consistent with higher clearance, these compounds had also considerably lower oral bioavailability after po administration at 10 mg/kg dose level being 10.06, 22.93, 42.19, and 43.74% for **17**, **11**, **15**, and **16**, respectively - comparing to 77.38% observed for **OATD-01**.

General

All solvents, substrates and reagents that were commercially available were used without further purification. TLC analysis was performed using pre-coated glass plates (0.2 ± 0.03 mm thickness, GF-254, particle size 0.01–0.04 mm) from Fluorochem Ltd, UK. Column chromatography was performed using high-purity grade silica gel (pore size 60 Å, 220-440 mesh particle size, 35-75 µm particle size) from Fluka. Preparative HPLC was performed on LC-20AP Shimadzu with ELSD-LTII detector equipped with Hypersil GOLD 21.2/250 mm, 5 µm C18 column. ^{1}H NMR spectra were recorded on Agilent Mercury 400 MHz spectrometer and on Bruker AVANCE DRX500, AVANCE DRX600 or Bruker AVANCE II PLUS (respectively at 500, 600 or 700 MHz) NMR spectrometers. All spectra were recorded in appropriate deuterated solvents ($CDCl_3$, DMSO-d_6, D_2O, Methanol-d_4, etc.) that were commercially available. Resonances are given in parts per million relative to tetramethylsilane. Data are reported as follows: chemical shift, multiplicity (s = singlet, d = doublet, t = triplet, m = multiplet, br = broad), coupling constants (J in Hz) and integration. LC-MS spectra were recorded on a Shimadzu LC-20AD LPG separation module with a SPD-M20A UV detector and LCMS-2020 mass detector equipped with Kinetex 2.1/50 mm, 2.6 µm C18 column eluted with 0.5 mL/min flow of 10-90 % gradient (over 6 min) of acetonitrile in water. Purities of all final reported compounds were greater than 95% based on HPLC chromatograms. For compounds **5** to **17** and compound **3** HPLC analyses were performed on a Shimadzu UPLC system fitted with a Phenomenex 3µm, 100A, C18(2) column (3 mm×150 mm) and with UV detection (200-400 nm), gradient 10−90% of acetonitrile in water, flow rate 1.5 mL/min over 10 min, then 100% acetonitrile over 4min. For compound **4** HPLC analyses were performed on a Waters Acquity UPLC system fitted with a Phenomenex 5µm, 100A, PFP(2) column (4.6 mm×150 mm) and with UV detection (220 nm), gradient 10−90% of acetonitrile in water, flow rate 1.5 mL/min over 10 min, then 90% acetonitrile over 5min. Purification of the final compounds by preparative HPLC was accomplished on C-18 250×21 mm column in 0.05% TFA in water / acetonitrile 95:5 → 45:55 gradient over 30 minutes followed by freeze-drying of the pooled fractions containing pure products. In some cases, the so obtained trifluoroacetate salts of the final compounds were judged to be of insufficient quality for biological testing due to their physical appearance (oils with a distinguished scent of trifluoroacetic acid). Therefore, they were re-dissolved in a small amount of 0.1 M HCl and subjected to the second lyophilization providing well-behaving hydrochloride salts.

Experimental procedures for compounds 5, 7, 8, 15 - 17.

General procedure A (reductive amination of *N*-Boc-4-piperidone with cyclic secondary amines).

To a stirred solution of N-Boc-4-piperidone (0.4 g, 1 mmol) and secondary amine or its hydrochloride salt (1 mmol) in 1,2-dichloroethane (3 mL), acetic acid (0.36 mL, 3 mmol) was added and the resulting mixture was stirred at room temperature until clear solution was obtained (overnight, if neccessary). It was concentrated *in vacuo* to about 30-50% of its initial volume and sodium triacetoxyborohydride (0.42 g, 2 mmol) was added. The thick reaction mixture was stirred at ambient temperature overnight after which time it was diluted with 3 mL of DCM and quenched by addition of 5% solution of $NaHCO_3$ (6 mL). Compounds were isolated by usual aqueous work-up and purified by flash chromatography in appropriate solvent system.

General procedure B (*N*-Boc-group removal followed by 3-aminotriazole ring formation).

To a solution of Boc-protected compounds in EtOAc (0.5 mL/mmol), 4 M HCl in EtOAc solution was added (3 mL/mmol) and reaction was stirred at room temperature until chromatography (either TLC or LC-MS) indicated complete consumption of a starting material (typically 30 min – 2 hours). Volatiles were removed *in vacuo* and the residue was triturated thrice with diethyl ether and dried under high vacuum for 1 hour.

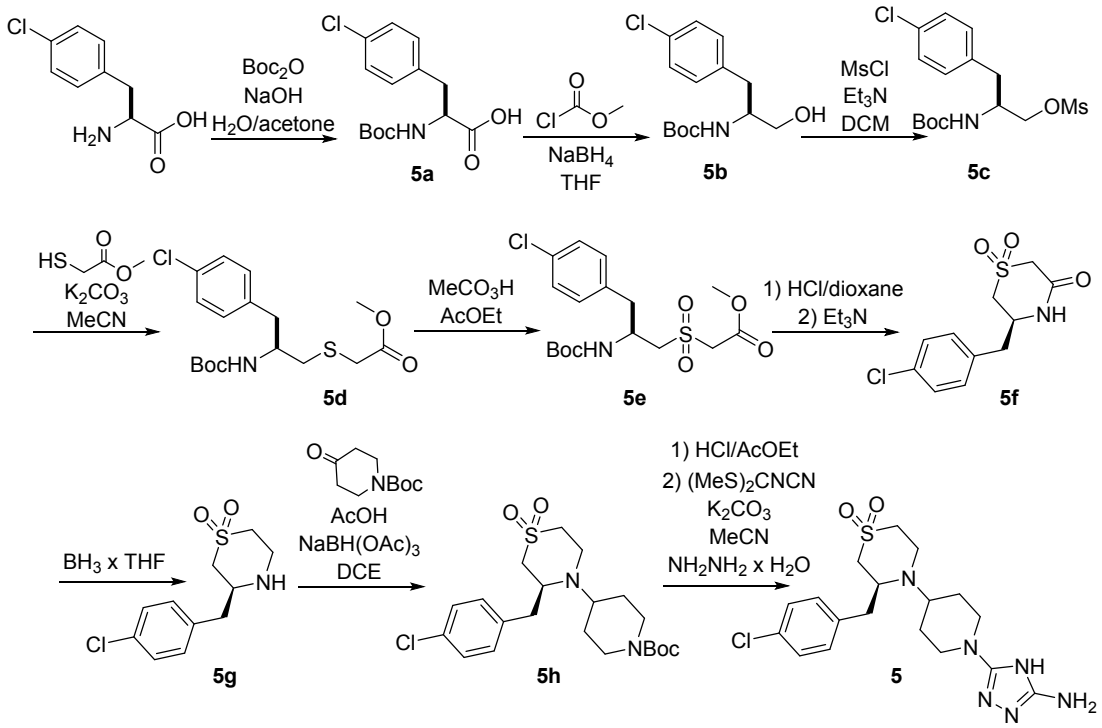
Or

The N-Boc protected amine was treated with solution of TFA (6 equivalents) DCM for the time necessary for complete consumption of the starting material (typically 30 minutes – 2 hours). The volatiles were then removed *in vacuo* providing de-protected amine in the form of its TFA salt.

The crude hydrochloride or TFA salt was suspended in acetonitrile (4 mL/mmol, assuming quantitative yield of the deprotection step) and solid K_2CO_3 (3 equivalents) followed by *S*,*S*`-dimethyl *N*-cyanodithioiminocarbonate (1.1 equivalent) were added and the reaction mixture was refluxed until complete consumption of the starting material was achieved as judged by chromatography (typically 2-3 hours). Hydrazine hydrate (4 equivalents) was then added and reaction was further refluxed for 3-5 hours, after which time it was cooled, and poured into the vigorously stirred biphasic mixture of 5% $NaHCO_3$ and ethyl acetate. After the separation of the phases, the organic layer was additionally washed with water and brine, dried over anhydrous $MgSO_4$, filtered and concentrated. Depending on the amount of the compound synthesized, the final products were purified by crystallization, flash chromatography or reversed-phase chromatography.

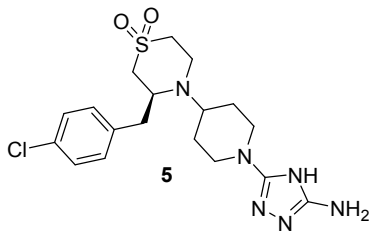
General procedure C (reduction of carbonyl group).

To the solution of either morpholin-3-one or 2-piperazinone or amide in THF (3 mL/mmol) borane-dimethylsulfide complex (BH_3 x DMS; 3 equivalents) is added and the reaction mixture is refluxed for 3 hours, after which time the TLC or LC-MS control shows complete consumption of the starting material. Reaction mixture is cooled to room temperature and 2 M HCl is cautiously added (6 equivalents with respect to the starting material). The resulting reaction mixture is refluxed for 2 hours and cooled back to room temperature. The pH of the solution is then adjusted to strongly alkaline (~10) by a dropwise addition of 6 M NaOH. The organic layer is separated and the aqueous layer is additionally extracted with diethyl ether. Combined organic extracts are then dried over $MgSO_4$, filtered and the solvents are evaporated. Crude product obtained is, in most cases sufficiently pure to be used in the next step without any additional purification.



Example 5.

Synthesis of (*S*)-4-(1-(5-amino-4*H*-1,2,4-triazol-3-yl)piperidin-4-yl)-3-(4-chlorobenzyl)thiomorpholine 1,1-dioxide (**5**).



Step 1.

Synthesis of (*S*)-2-((*tert*-butoxycarbonyl)amino)-3-(4-chlorophenyl)propanoic acid (**5a**).

To a solution of *p*-chloro-L-phenylalanine (18.0 g, 75 mmol) in acetone-water (150 mL : 150 mL) was added sodium hydroxide (6 g, 150 mmol) at 0°C followed by di-*tert*-butyl dicarbonate (16.4 g, 75 mmol). The reaction mixture was stirred at room temperature overnight. Acetone was evaporated. Aqueous layer was acidified to pH 2 with 2 M HCl and extracted with ethyl acetate. Organic layer was dried over $MgSO_4$, filtered and concentrated *in vacuo*. The crude product was crystallized from hexane to obtain **5a** as a white solid in 80% yield (18.0 g; 60 mmol).

ESI-MS m/z for $C_{14}H_{19}ClNO_4$ found 299.8/301.8 $[M+H]^+$; ^1H NMR (500 MHz, DMSO-d_6) δ: 7.29 (d, *J* = 8.3 Hz, 2H), 7.21 (d, *J* = 8.3 Hz, 2H), 7.02 (d, *J* = 7.3 Hz, 1H), 4.08-3.99 (m, 1H), 2.96 (dd, *J* = 4.3, 13.7Hz, 1H), 2.76 (dd, *J* = 10.5, 13.6 Hz, 1H).

Step 2.

Synthesis of *tert*-butyl (*S*)-(1-(4-chlorophenyl)-3-hydroxypropan-2-yl)carbamate (**5b**).

The Boc-L-*p*-chlorophenylalanine (14 g; 46.72 mmol) was dissolved in THF (187 ml) and *N*-methylmorpholine (6.2 mL; 56.1 mmol) was added. The solution was cooled to –15ºC and methyl chloroformate (4.2 mL; 56.1 mmol) was added and the mixture was stirred for additional 20 minutes. The precipitate was filtered-off and filtrate was transferred into a larger round bottomed flask. The suspension of $NaBH_4$ (44 g; 116.8 mmol) in water (46 mL) was then cautiously added (Caution: intense foaming!) and the reaction mixture was allowed to stir at room temperature overnight. 1 M NaOH was added in volume equal to that of THF used and the mixture was stirred for additional 30 minutes after which time it was extracted with ethyl acetate (3×). The organic phase was dried over $MgSO_4$, filtered and concentrated *in vacuo* and the crude product was used to the next step without additional purification. Compound **5b** was obtained in 60% yield (8 g; 28.06 mmol).

ESI-MS m/z for $C_{14}H_{21}ClNO_3$ found 285.9/287.9 $[M+H]^+$

Step 3.

Synthesis of (*S*)-2-((*tert*-butoxycarbonyl)amino)-3-(4-chlorophenyl)propyl methanesulfonate (**5c**).

To a solution of substrate **5b** (1.8 g; 6.29 mmol) and triethylamine (1.4 mL; 9.44 mmol) in dichloromethane (20 mL), mesyl chloride (0.73 mL; 9.44 mmol) was added dropwise. After 1 hour of stirring, reaction mixture was diluted with dichloromethane, washed with 2 M HCl, 5% aq.$NaHCO_3$, brine, dried and concentrated *in vacuo*. The residue was washed with ether and the compound **5c** was obtained as a white solid in 92% yield (2.09 g; 5.76 mmol).

ESI-MS m/z for $C_{15}H_{22}ClNO_5SNa$ found 386.1/388.1 $[M+Na]^+$

Step 4.

Synthesis of methyl (*S*)-2-((2-((*tert*-butoxycarbonyl)amino)-3-(4-chlorophenyl)propyl)thio)acetate (**5d**).

Mesylate **5c** (2.09 g; 5.76 mmol), K_2CO_3 (1.58 g; 11.48 mmol) and methyl thioglycolate (0.65 mL; 11.48 mmol) in acetonitrile (20 mL) were heated under reflux for 30 minutes, then the reaction mixture was diluted with water and product was extracted with diethyl ether. The organics were washed with 2 M HCl, 5% aq. $NaHCO_3$, brine, dried and concentrated *in vacuo* and the crude product was used to the next step without additional purification. Compound **5d** was obtained in 98% yield (2.1 g; 5.63 mmol).

ESI-MS m/z for $C_{17}H_{25}ClNO_4S$ found 374.1/376.1 $[M+H]^+$

Step 5.

Synthesis of methyl (*S*)-2-((2-((*tert*-butoxycarbonyl)amino)-3-(4-chlorophenyl)propyl)sulfonyl)acetate (**5e**).

To a solution of **5d** (2.1 g; 5.63 mmol) in ethyl acetate, peracetic acid (2.1 mL; 12.83 mmol; 39% in AcOH) was added dropwise, then the reaction was stirred overnight at room temperature and concentrated to dryness. The residue was triturated with diethyl ether and crude product was used in the next step. Compound **5e** was obtained in 99% yield (2.26 g; 5.57 mmol).

ESI-MS m/z for $C_{17}H_{25}ClNO_6S$ found 406.1/408.1 $[M+H]^+$

Step 6.

Synthesis of (*S*)-5-(4-chlorobenzyl)thiomorpholin-3-one 1,1-dioxide (**5f**).

Compound **5e** (2.26 g; 5.57 mmol) was treated with HCl/dioxane and stirred for 1 hour, then concentrated to dryness. The residue was dissolved in MeOH and treated with Et_3N (1.8 mL; 11.48 mmol). After 30 minutes the reaction was concentrated, the residue was taken into dichloromethane, washed with 2 M HCl, 5% aq. $NaHCO_3$, brine, dried over anhydrous $MgSO_4$ and concentrated providing orange solid. Colored impurities were removed by trituration with diethyl ether. Compound **5f** was obtained as a white solid in 34% yield (0.52 g; 1.9 mmol).

ESI-MS m/z for $C_{11}H_{13}ClNO_3S$ found 274.1/276.1 $[M+H]^+$; ^1H NMR (500 MHz, DMSO-d_6) δ 8.43 (bs, 1H), 7.35 ($J_{AA'BB'}$ = 8.3 Hz, 2H), 7.26 ($J_{AA'BB'}$ = 8.3 Hz, 2H), 4.17-4.11 (m, 1H), 4.00 (dd, *J* = 2.6, 16.1 Hz, 1H), 3.97-3.90 (m, 1H), 3.23-3.17 (m, 1H), 3.09 (dd, *J* = 11.1, 13.7 Hz, 1H), 2.97 (dd, *J* = 4.7, 13.5 Hz, 1H), 2.82 (dd, *J* = 7.5, 13.5 Hz, 1H).

Step 7.

Synthesis of (*S*)-3-(4-chlorobenzyl)thiomorpholine 1,1-dioxide (**5g**).

Compound **5f** (0.42 g; 1.53 mmol) was dissolved in dry THF (15 mL) and borane-tetrahydrofurane complex (4.6 mL; 4.6 mmol) was carefully added and the reaction was heated with stirring for 1 hours. After this time the TLC revealed the complete consumption of the starting material. The reaction mixture was carefully quenched with water. 1 N NaOH was added and the reaction mixture was extracted with diethyl ether. Organics were dried over $MgSO_4$ and concentrated *in vacuo* and the crude product was used to the next step without additional purification. Compound **5g** was obtained in 99% yield (391 mg; 1.51 mmol).

ESI-MS m/z for $C_{11}H_{15}ClNO_2S$ found 260.1/262.1 $[M+1]^+$; ^1H NMR (500 MHz, DMSO-d_6) δ 7.31 ($J_{AA'BB'}$ = 8.3 Hz, 2H), 7.21 ($J_{AA'BB'}$ = 8.3 Hz, 2H), 3.37-3.17 (m, 2H), 3.09-3.00 (m, 1H), 2.97-2.74 (m, 4H), 2.72-2.63 (m, 2H).

Step 8.

Synthesis of *tert*-butyl (*S*)-4-(3-(4-chlorobenzyl)-1,1-dioxidothiomorpholino)piperidine-1-carboxylate (**5h**).

The title compound (**5h**) was obtained according to the General Procedure A from compound **5g** (391 mg; 1.51 mmol) in 91% yield (610 mg; 1.38 mmol).

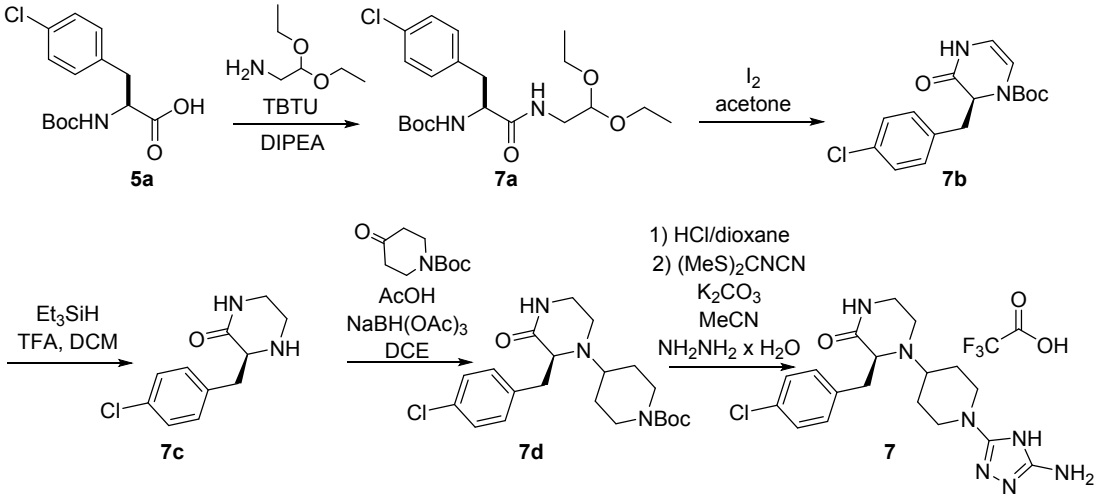
ESI-MS m/z for $C_{21}H_{32}ClN_2O_4S$ found 443.1/445.1 $[M+H]^+$; ^1H NMR (500 MHz, $CDCl_3$) δ 7.24 ($J_{AA'BB'}$ = 8.3 Hz, 2H), 7.10 ($J_{AA'BB'}$ = 8.3 Hz, 2H), 3.85-3.72 (m, 2H), 3.52-3.42 (m, 1H), 3.41-3.32 (m, 1H), 3.14-3.01 (m, 2H), 3.02-2.57 (m, 6H), 2.26-1.98 (m, 2H), 1.84-1.73 (m, 2H), 1.73-1.63 (m, 2H), 1.42 (s, 9H).

Step 9.

Synthesis of (*S*)-4-(1-(5-amino-4*H*-1,2,4-triazol-3-yl)piperidin-4-yl)-3-(4-chlorobenzyl)thiomorpholine 1,1-dioxide (**5**).

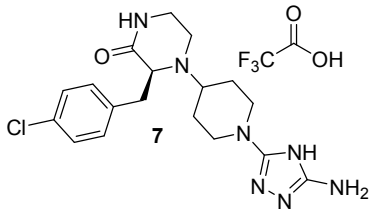
The title compound (**5**) was obtained according to the General Procedure B from compound **5h** (270 mg; 0.61 mmol) in 30% yield (75 mg; 0.18 mmol).

^1H NMR (500 MHz, DMSO-d_6) δ 7.31 ($J_{AA'BB'}$ = 8.5 Hz, 2H), 7.24 ($J_{AA'BB'}$ = 8.5 Hz, 2H), 5.48 (bs, 2H), 3.71-3.64 (m, 2H), 3.52-3.43 (m, 1H), 3.11-2.96 (m, 3H), 2.94-2.79 (m, 4H), 2.78-2.70 (m, 1H), 2.64-2.52 (m, 2H), 1.65-1.55 (m, 2H), 1.37-1.18 (m, 3H); ^1H NMR (700 MHz, DMSO-d_6 + D_2O, 348 K) δ 7.32 – 7.29 (m, 2H), 7.29 – 7.20 (m, 2H), 3.79 – 3.63 (m, 2H), 3.63 – 3.52 (m, 1H), 3.40 – 3.31 (m, 1H), 3.19 – 3.12 (m, 1H), 3.06 – 2.99 (m, 2H), 2.99 – 2.76 (m, 5H), 2.72 – 2.60 (m, 2H), 1.69 – 1.61 (m, 2H), 1.45 – 1.25 (m, 2H); ^{13}C NMR (176 MHz, DMSO-d_6) δ 138.3, 131.7 (2×), 131.3, 128.7 (2×), 60.4, 57.5, 55.8, 52.2, 50.2, 46.0, 45.9, 41.5, 35.2, 30.2, 28.0, 21.2; HRMS (ESI) *m/z* calc. for $C_{18}H_{26}ClN_6O_2S$ $[M+H]^+$ 425.1521 found 425.1533.



Example 7.

Synthesis of (*S*)-4-(1-(5-amino-4*H*-1,2,4-triazol-3-yl)piperidin-4-yl)-3-(4-chlorobenzyl)piperazin-2-one 2,2,2-trifluoroacetate (**7**).



Step 1.

Synthesis of *tert*-butyl (*S*)-(3-(4-chlorophenyl)-1-((2,2-diethoxyethyl)amino)-1-oxopropan-2-yl)carbamate (**7a**).

Compound **5a** (1.00 g; 3.34 mmol) was dissolved in DCM (13.4 mL) and diisopropylethylamine (0.87 mL; 5.0 mmol) was added at room temperature followed by addition of aminoacetaldehyde diethylacetal (0.54 mL; 3.67 mmol) and O-(benzotriazol-1-yl)-*N*,*N*,*N'*,*N'*-tetramethyluronium tetrafluoroborate (TBTU) (1.13 g; 3.50 mmol). The reaction mixture was stirred for 3 hours at room temperature, diluted with methylene chloride and washed with 1 M K_2CO_{3aq} and 1 M HCl_{aq}, brine, dried over anhydrous $MgSO_4$ filtered and concentrated *in vacuo*. The residue was purified by flash chromatography (DCM/MeOH 100:0 to 100:1 *v*/*v*) and **7a** was obtained in 92% yield (1.27 g; 3.07 mmol).

ESI-MS m/z for $C_{20}H_{32}ClN_2O_5$ found 415.4/417.4 $[M+H]^+$; ^1H NMR (500 MHz, $CDCl_3$) δ 7.23 (m, 2H), 7.11 (m, 2H), 5.92 (brs, 1H), 5.02 (brs, 1H), 4.28 (brs, 2H), 3.64-3.56 (m, 2H), 3.47-3.43 (m, 1H), 3.40-3.30 (m, 2H), 3.25 (brs, 1H), 2.99 (brs, 2H), 1.38 (s, 9H), 1.14 (q, *J* = 6.9Hz, 6H).

Step 2.

Synthesis of *tert*-butyl (*S*)-2-(4-chlorobenzyl)-3-oxo-3,4-dihydropyrazine-1(2*H*)-carboxylate (**7b**).

Compound **7a** (1.38 g; 3.33 mmol) was dissolved in acetone (33 mL) and then I_2 (85 mg; 0.33 mmol) was added and the mixture was stirred overnight at room temperature. The solvent was removed *in vacuo* and oily residue was dissolved in Et_2O, then washed twice with 10 % $Na_2S_2O_4$. The organic layer was dried over $MgSO_4$, filtered and concentrated *in vacuo*. The residue was purified by flash chromatography (DCM/MeOH 100:0 to 100:1 *v*/*v*) and **7b** was obtained in 82% yield (0.88 g; 2.73 mmol).

ESI-MS m/z for $C_{16}H_{20}ClN_2O_3$ found 322.7/324.7 $[M+H]^+$; ^1H NMR (500 MHz, $CDCl_3$) – two conformers present due to hindered rotation δ {[7,62 (1^{st} isomer, brs), 7.58 (2^{nd} isomer, brs)], 1H}, {[7.25 (2^{nd} isomer, d, *J* = 8.0Hz), 7.20 (1^{st} isomer, d, *J* = 8.0Hz)], 2H}, {[7,08 (1^{st} isomer, d, *J* = 8.0Hz), 7.06 (2^{nd} isomer, d, *J* = 8Hz)], 2H}, {[6.36 (2^{nd} isomer, d, *J* = 5.8 Hz), 6.10 (1^{st} isomer, d, *J* = 5.8 Hz)], 1H}, {[5.67 (2^{nd} isomer, t, *J* = 5.1 Hz), 5.42 (1^{st} isomer, t, *J* = 5.1 Hz)], 1H}, {[4.99-4.95 (1^{st} isomer, m), 4.80-4.76 (2^{nd} isomer, m)],

1H}, {[3.02-2.96 (1st isomer, m), 2.90-2.86 (2nd isomer, m)], 2H}, {[1.35(1st isomer, s), 1.17 (2nd isomer, s)], 9H}.

Step 3.

Synthesis of (*S*)-3-(4-chlorobenzyl)piperazin-2-one (**7c**).

To a solution of compound **7b** (0.58 g; 1.80 mmol) in dichloromethane (DCM) (5 mL), triethylsilane Et_3SiH (1.4 mL; 8.9 mmol) was added followed by slow addition of trifluoroacetic acid (TFA) (1.3 mL; 17.8 mmol) and the reaction mixture was stirred overnight at room temperature. The volatiles were then removed *in vacuo* and the residue was taken between 1 M NaOH and DCM. The organic phase was washed with brine, dried over $MgSO_4$, filtered and concentrated *in vacuo* and the crude product was used to the next step without additional purification. Compound **7c** was obtained in 79% yield (0.32 g; 1.43 mmol).

ESI-MS m/z for $C_{11}H_{14}ClN_2O$ found 225.2/ 227.2 $[M+1]^+$; ^{1}H NMR (500 MHz, $CDCl_3$) δ 7.29-7.23 (m, 2H), 7.18 (d, *J* = 8.3 Hz, 2H), 6.11 (s, 1H), 3.59 (dd, *J* = 9.6, 3.6 Hz, 1H), 3.41-3.30 (m, 2H), 3.24 (dq, *J* = 11.2, 3.6 Hz, 1H), 3.06 (dt, *J* = 12.6, 3.9 Hz, 1H), 2.93-2.82 (m, 2H).

Step 4.

Synthesis of (*S*)-*tert*-butyl 4-(2-(4-chlorobenzyl)-3-oxopiperazin-1-yl)piperidine-1-carboxylate (**7d**).

The title compound (**7d**) was obtained according to the General Procedure A from compound **7c** (278 mg; 1.24 mmol) in 60% yield (300 mg; 0.74 mmol).

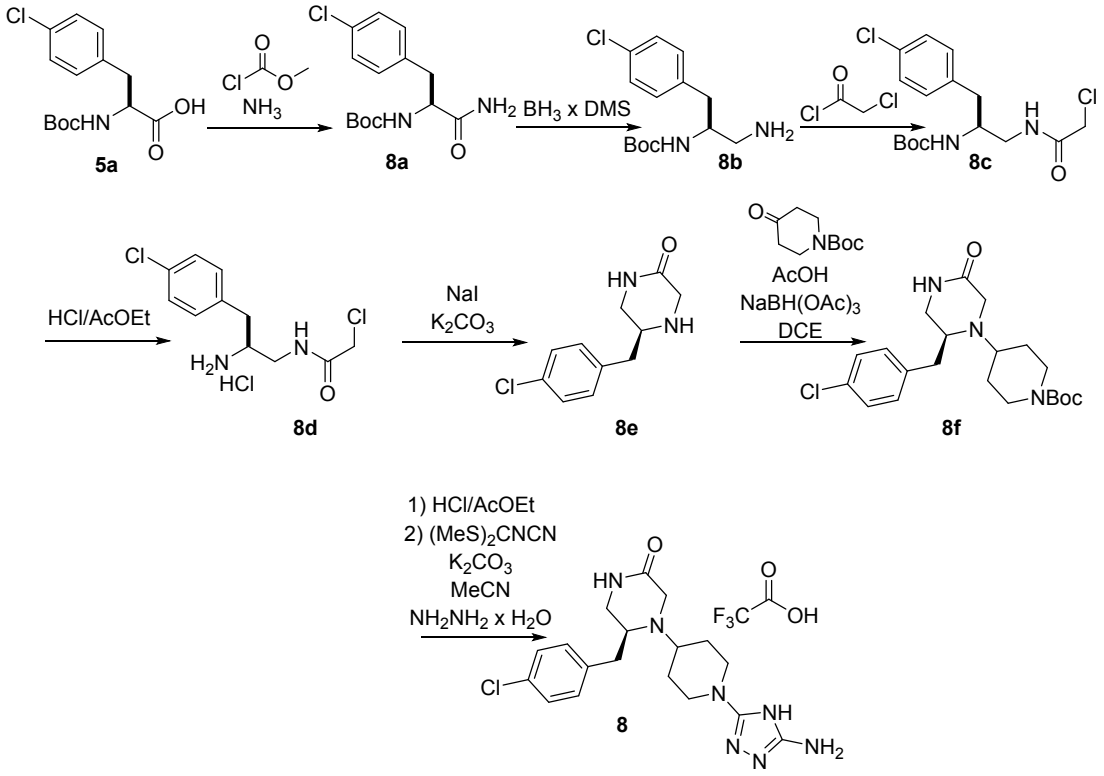
ESI-MS m/z for $C_{21}H_{31}ClN_3O_3$ found 408.2/410.2 $[M+H]^+$; ^{1}H NMR (500 MHz, $CDCl_3$) δ 7.28 – 7.13 (m, 4H), 6.29 (s, 1H), 4.27 – 3.95 (m, 2H), 3.66 – 3.47 (m, 1H), 3.26 – 3.12 (m, 1H), 3.12 – 3.02 (m, 2H), 3.04 – 2.88 (m, 2H), 2.79 – 2.57 (m, 3H), 2.09 – 1.99 (m, 1H), 1.70 – 1.53 (m, 2H), 1.52 – 1.38 (m, 9H), 1.33 – 1.24 (m, 2H).

Step 5.

Synthesis of (*S*)-4-(1-(5-amino-4*H*-1,2,4-triazol-3-yl)piperidin-4-yl)-3-(4-chlorobenzyl)piperazin-2-one 2,2,2-trifluoroacetate (**7**).

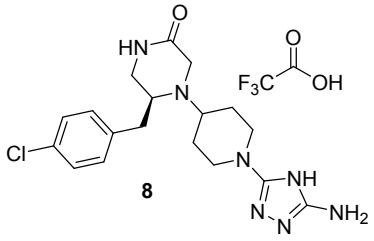
The title compound (**7**) was obtained as a TFA salt according to the General Procedure B from compound **7d** (300 mg; 0.74 mmol) in 41% yield (150 mg; 0.3 mmol).

^{1}H NMR (500 MHz, DMSO-d_6) δ 7.88 (s, 1H), 7.32-7.22 (m, 4H), 3.85-3.59 (m, 4H), 3.15-2.75 (m, 8H), 1.73 (d, *J* = 12.2 Hz, 1H), 1.66-1.53 (m, 1H), 1.51-1.28 (m, 2H); ^{1}H NMR (700 MHz, DMSO-d_6 + D_2O, 348 K) δ 7.34 – 7.20 (m, 4H), 3.73 – 3.55 (m, 3H), 3.24 – 3.17 (m, 1H), 3.11 – 3.01 (m, 3H), 2.99 – 2.76 (m, 5H), 1.75 – 1.62 (m, 2H), 1.51 – 1.33 (m, 2H); ^{13}C NMR (176 MHz, DMSO-d_6) δ 159.4, 137.4, 131.9 (2×), 131.5, 128.3 (2×), 62.0, 57.2, 45.7, 45.6, 41.3, 40.0, 38.5, 35.1, 28.3, 27.0, 25.2; HRMS (ESI) *m/z* calc. for $C_{18}H_{25}ClN_7O$ $[M+H]^+$ 390.1804 found 390.1809.



Example 8.

Synthesis of (*S*)-4-(1-(5-amino-4*H*-1,2,4-triazol-3-yl)piperidin-4-yl)-5-(4-chlorobenzyl)piperazin-2-one 2,2,2-trifluoroacetate (**8**).



Step 1.

Synthesis of *tert*-butyl (*S*)-(1-amino-3-(4-chlorophenyl)-1-oxopropan-2-yl)carbamate (**8a**).

The Boc-L-*p*-chlorophenylalanine **5a** (1.4 g; 4.67 mmol) was dissolved in dichloromethane (DCM) (40 ml) and *N*-methylmorpholine (0.62 mL; 5.61 mmol) was added. The solution was cooled to –15ºC and methyl chloroformate (0.42 mL; 5.61 mmol) was added dropwise and the mixture was stirred for additional 30 minutes at which time aqueous ammonia (28% in H_20; 0.5 mL; 13.38 mmol) was added. The white precipitate was formed in all volume of reaction. The precipitate was filtered-off and washed with DCM, aqueous ammonia and ether. The phases was separated and an organic one was dried over $MgSO_4$, filtered and concentrated *in vacuo* and the crude product was used to the next step without additional purification. Compound **8a** was obtained as a white powder in 98% yield (1.36 g; 4.56 mmol). ESI-MS m/z for $C_{14}H_{20}ClN_2O_3$ found 299.0/301.0 $[M+H]^+$, 321.0/323.0 $[M+Na]^+$

Step 2.

Synthesis of *tert*-butyl (*S*)-(1-amino-3-(4-chlorophenyl)propan-2-yl)carbamate (**8b**).

The title compound (**8b**) was prepared according to the General Procedure C from compound **8a** (1.36 g; 4.56 mmol) in 28% yield (0.36 g; 1.27 mmol).

ESI-MS m/z for $C_{14}H_{22}ClN_2O_2$ found 285.3/3287.3 $[M+H]^+$

Step 3.

Synthesis of *tert*-butyl (*S*)-(1-(2-chloroacetamido)-3-(4-chlorophenyl)propan-2-yl)carbamate (**8c**).

To a solution of **8b** (0.36 g; 1.27 mmol) in DCM (20 mL) chloroacetyl chloride (0.1 mL; 1.33 mmol) was added followed by Et_3N (0.26 mL; 1.9 mmol) and the reaction mixture was stirred at ambient temperature for 15 minutes. The reaction progress was monitored by TLC and LC-MS. When analysis indicated completion of the reaction, the mixture was diluted with water and extracted with AcOEt. Combined organic solutions were washed with 2 M HCl (2×), water and brine and dried over $MgSO_4$, filtered and concentrated *in vacuo* and the crude product was used to the next step without additional purification. Compound **8c** was obtained in 99% yield (0.45 g; 1.26 mmol).

ESI-MS m/z for $C_{16}H_{23}Cl_2N_2O_3$ found 361.3/363.3 $[M+H]^+$, 383.3/385.3 $[M+Na]^+$

Step 4.

Synthesis of (*S*)-*N*-(2-amino-3-(4-chlorophenyl)propyl)-2-chloroacetamide hydrochloride (**8d**).

Boc-protected compound **8c** (0.45 g; 1.26 mmol) was treated with 4M HCl in EtOAc solution (5 mL) and reaction was stirred in room temperature overnight. Volatiles were removed *in vacuo* and **8d** was obtained as a hydrochloride salt in 95% yield (355 mg; 1.2 mmol).

ESI-MS m/z for $C_{11}H_{15}Cl_2N_2O$ found 261.2/263.2 $[M+H]^+$

Step 5.

Synthesis of (*S*)-5-(4-chlorobenzyl)piperazin-2-one (**8e**).

The compound **8d** (355 mg; 1.2 mmol) was dissolved in acetonitrile (50 mL) and to this solution anhydrous potassium carbonate (437 mg; 3.16 mmol) and sodium iodide (20 mg; 1.33 mmol) were sequentially added. The reaction was stirred at 50°C for 5 hours and then at room temperature overnight. The solids were filtered off, the filtrate was concentrated and the product was purified by flash column chromatography (DCM/MeOH 40:1 to 5:1 *v*/*v*) and **8e** was obtained in 59% yield (160 mg; 0.71 mmol).

ESI-MS m/z for $C_{11}H_{14}ClN_2O$ found 225.2/227.2 $[M+H]^+$

Step 6.

Synthesis of *tert*-butyl (*S*)-4-(2-(4-chlorobenzyl)-5-oxopiperazin-1-yl)piperidine-1-carboxylate (**8f**).

The title compound (**8f**) was obtained according to the General Procedure A from compound **8e** (160 mg; 0.71 mmol) in 61% yield (175 mg; 0.43 mmol).

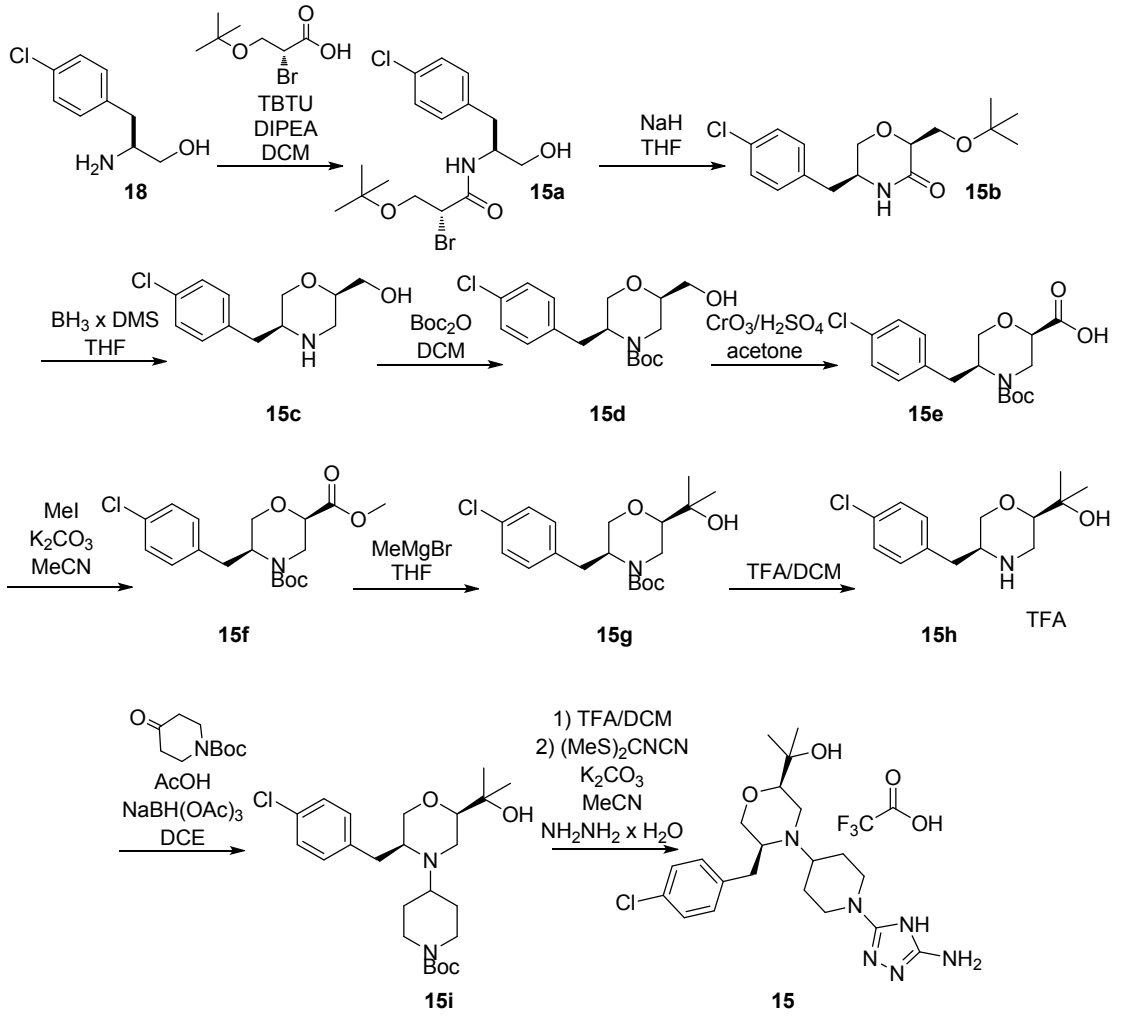
ESI-MS m/z for $C_{21}H_{31}ClN_3O_3$ found 408.4/410.4 $[M+H]^+$

Step 7.

Synthesis of (*S*)-4-(1-(5-amino-4*H*-1,2,4-triazol-3-yl)piperidin-4-yl)-5-(4-chlorobenzyl)piperazin-2-one 2,2,2-trifluoroacetate (**8**).

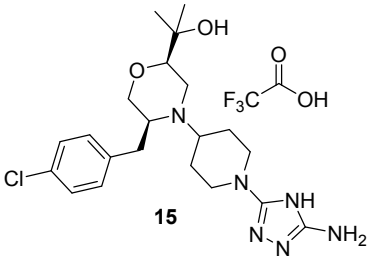
The title compound (**8**) was obtained as a TFA salt according to the General Procedure B from compound **8f** (175 mg; 0.43 mmol) in 19% yield (42 mg; 0.083 mmol).

^{1}H NMR (500 MHz, D_2O) δ 7.28 ($J_{AA'BB'}$ = 7.3 Hz, 2H), 7.14 ($J_{AA'BB'}$ = 7.7 Hz, 2H), 4.07-4.12 (m, 1H), 4.01 (d, *J* = 16.7 Hz, 1H), 3.86 (d, *J* = 16.9 Hz, 1H), 3.73-3.77 (m, 2H), 3.32-3.36 (m, 1H), 3.17-3.22 (m, 2H), 2.81-3.04 (m, 4H), 2.09-2.15 (m, 2H), 1.68-1.75 (m, 2H); ^{1}H NMR (700 MHz, DMSO-d_6 + D_2O, 348 K) δ 7.41 – 7.24 (m, 4H), 3.87 – 3.77 (m, 2H), 3.74 – 3.69 (m, 1H), 3.67 – 3.62 (m, 1H), 3.60 – 3.52 (m, 1H), 3.37 – 3.23 (m, 3H), 3.12 – 3.07 (m, 1H), 3.06 – 2.99 (m, 1H), 2.99 – 2.86 (m, 2H), 2.82 – 2.72 (m, 1H), 2.02 – 1.91 (m, 2H), 1.71 – 1.56 (m, 2H); ^{13}C NMR (176 MHz, DMSO-d_6) δ 159.2, 135.0, 132.4, 131.7 (2×), 129.2 (2×), 59.2, 54.3, 48.1, 47.4, 45.3, 45.1, 40.3, 40.0, 26.4, 25.4, 24.5; HRMS (ESI) *m/z* calc. for $C_{18}H_{25}ClN_7O$ $[M+H]^+$ 390.1804 found 390.1821.



Example 15.

Synthesis of 2-((2*R*,5*S*)-4-(1-(5-amino-4*H*-1,2,4-triazol-3-yl)piperidin-4-yl)-5-(4-chlorobenzyl)morpholin-2-yl)propan-2-ol 2,2,2-trifluoroacetate (**15**).



15

Step 1.

Synthesis of (*R*)-2-bromo-3-(*tert*-butoxy)-*N*-((*S*)-1-(4-chlorophenyl)-3-hydroxypropan-2-yl propanamide (**15a**).

To the solution of (*S*)-2-amino-3-(4-chlorophenyl)propan-1-ol (**18**) (6 g; 32.14 mmol) and (2*R*)-2-bromo-3-*tert*-butoxypropanoic acid (7.23 g; 32.14 mmol) in DCM (100 mL) diisopropylethylamine (DIPEA; 8.18 mL; 48.21 mmol) and TBTU (10.32 g; 32.14 mmol) were added sequentially and the reaction mixture was stirred at room temperature overnight. After this time TLC control showed complete consumption of the starting materials so the reaction mixture was transferred to the separatory funnel and washed sequentially with 1 M HCl, 1 M K_2CO_3. The organic phase was dried over $MgSO_4$, filtered and concentrated *in vacuo* and the crude product was purified by flash column chromatography (DCM/MeOH 100:1 *v*/*v*) and **15a** was obtained as a colorless oil in 88% yield (11 g; 28.13 mmol).

ESI-MS m/z for $C_{16}H_{23}BrClNO_3Na$ found 414.3/416.3 $[M+Na]^+$

Step 2.

Synthesis of (2*S*,5*S*)-2-(*tert*-butoxymethyl)-5-(4-chlorobenzyl)morpholin-3-one (**15b**).

To the solution of **15a** (11 g; 28.13 mmol) in THF (280 mL) sodium hydride (NaH) (60% in mineral oil; 3.97 g; 99.31 mmol) was added in one portion and stirred at room temperature for 2 hours. The excess of NaH was then carefully quenched by dropwise addition of 2 M HCl to pH 3 and then additional volume of brine was added. The organic layer was separated and the aqueous layer was additionally extracted with diethyl ether. Combined organic extracts were then dried over $MgSO_4$, filtered and concentrated *in vacuo* and the crude product was purified by flash column chromatography (DCM/MeOH 100:1 *v*/*v*) and **15b** was obtained as a white solid in 91% yield (8 g; 25.71 mmol).

ESI-MS m/z for $C_{16}H_{22}ClNO_3Na$ found 334.1/336.1 $[M+Na]^+$

Step 3.

Synthesis of ((2*R*,5*S*)-5-(4-chlorobenzyl)morpholin-2-yl)methanol (**15c**).

The title compound (**15c**) was prepared according to the General Procedure C from compound **15b** (8 g; 25.71 mmol) in 77% yield (4.75 g; 19.7 mmol).

ESI-MS m/z for $C_{12}H_{17}ClNO_2$ found 242.2/244.2 $[M+H]^+$

Step 4.

Synthesis of *tert*-butyl (2*R*,5*S*)-5-(4-chlorobenzyl)-2-(hydroxymethyl)morpholine-4-carboxylate (**15d**).

To a solution of amino alcohol **15c** (2.87 g; 11.9 mmol) in dichloromethane (110 mL), di-*tert*-butyl dicarbonate (Boc_2O) (2.46 g; 11.3 mmol) was added and the reaction mixture was stirred at room temperature for 2 hours, after which time TLC showed almost complete consumption of the starting material. Volatiles were removed *in vacuo* and the residue was purified by column chromatography (hexane/AcOEt 1:1 *v*/*v*) and **15d** was obtained as a colorless oil in 77% yield (3.14 g; 9.2 mmol).

ESI-MS m/z for $C_{12}H_{17}ClNO_2$ found 242.1/246.1 $[M+H\text{-}Boc]^+$

Step 5.

Synthesis of (2*R*,5*S*)-4-(*tert*-butoxycarbonyl)-5-(4-chlorobenzyl)morpholine-2-carboxylic acid (**15e**).

To a cooled to 0°C solution of alcohol **15d** (1.8 g; 5.26 mmol) in acetone (40 mL), Jones reagent (12 mL; 2.6 M) was added dropwise. The reaction mixture was stirred at 0°C for 1 hour, and then isopropanol (*i*PrOH) (5 mL) was added. After 10 minutes ethyl acetate (150 mL) was added and the mixture was filtered through a pad of Celite. The filtrate was washed with brine, dried over $MgSO_4$ and evaporated affording the title compound **15e** as white foam in 91% yield (1.7 g; 4.79 mmol).

ESI-MS m/z for $C_{17}H_{21}ClNO_5Na$ found 378.3/380.3 $[M\text{-}H+Na]^+$, 256.1/258.1 $[M+H\text{-}Boc]^+$

Step 6.

Synthesis of 4-*tert*-butyl (2*R*,5*S*)-2-methyl 5-(4-chlorobenzyl)morpholine-2,4-dicarboxylate (**15f**).

To a solution of Boc-protected amino acid **15e** (1 g; 2.81 mmol) in acetonitrile (10 mL), potassium carbonate (0.77 g; 5.62 mmol) was added followed by methyl iodide (MeI) (0.26 mL; 4.21 mmol) at room temperature. After reaction was completed as judged by TLC, the reaction mixture was filtered and the solvent was evaporated. The residue was dissolved in ethyl acetate, washed with brine and dried over $MgSO_4$. The solvent was evaporated *in vacuo* and the crude product was used to the next step without additional purification. Compound **15f** was obtained as a yellow oil in 38% yield (0.4 g; 1.08 mmol).

ESI-MS m/z for $C_{18}H_{24}ClNO_5Na$ found 392.1/394.1 $[M+Na]^+$

Step 7.

Synthesis of *tert*-butyl (2*R*,5*S*)-5-(4-chlorobenzyl)-2-(2-hydroxypropan-2-yl)morpholine-4-carboxylate (**15g**).

To a solution of ester **15f** (0.4 g; 1.08 mmol) in dry THF (4 mL), solution of methylmagnesium bromide (1.1 mL; 3.24 mmol; 3 M in Et_2O) was added dropwise at room temperature. After 10 minutes the reaction mixture was quenched with saturated solution of ammonium chloride and extracted with ether. Organic phase was washed with brine, dried over $MgSO_4$ and concentrated *in vacuo* and the

crude product was used to the next step without additional purification. Compound **15g** was obtained in 99% yield (395 mg; 1.07 mmol).

ESI-MS m/z for $C_{19}H_{29}ClNO_4Na$ found 392.1/394.1 $[M+Na]^+$, 270.0/272.0 $[M+H\text{-}Boc]^+$

Step 8.

Synthesis of 2-((2*R*,5*S*)-5-(4-chlorobenzyl)morpholin-2-yl)propan-2-ol 2,2,2-trifluoroacetate (**15h**).

The compound **15g** (395 mg; 1.07 mmol) was treated with 3 mL of 50% trifluoroacetic acid (TFA) in dichloromethane for 30 minutes at room temperature, after which time the volatiles were removed *in vacuo* and the crude product was used to the next step without additional purification. Compound **15h** was obtained in 90% yield (0.37 g; 0.96 mmol).

ESI-MS m/z for $C_{14}H_{21}ClNO_2$ found 270.1/272.1 $[M+H]^+$; ^{1}H NMR (500 MHz, DMSO-d_6 + D_2O) δ 7.34 ($J_{AA'BB'}$ = 6.2 Hz, 2H), 7.25 ($J_{AA'BB'}$ = 6.4 Hz, 2H), 3.65-3.55 (m, 2H), 3.52-3.44 (m, 1H), 2.42-3.36 (m, 1H), 3.15-3.04 (m, 2H), 2.94-2.87 (m, 1H), 2.48 (m, 3H), 1.10 (s, 3H), 1.08 (s, 3H).

Step 9.

Synthesis of *tert*-butyl 4-((2*R*,5*S*)-5-(4-chlorobenzyl)-2-(2-hydroxypropan-2-yl)morpholino)piperidine-1-carboxylate (**15i**).

The title compound (**15i**) was obtained according to the General Procedure A from compound **15h** (0.37 g; 0.96 mmol) in 79% yield (344 mg; 0.76 mmol).

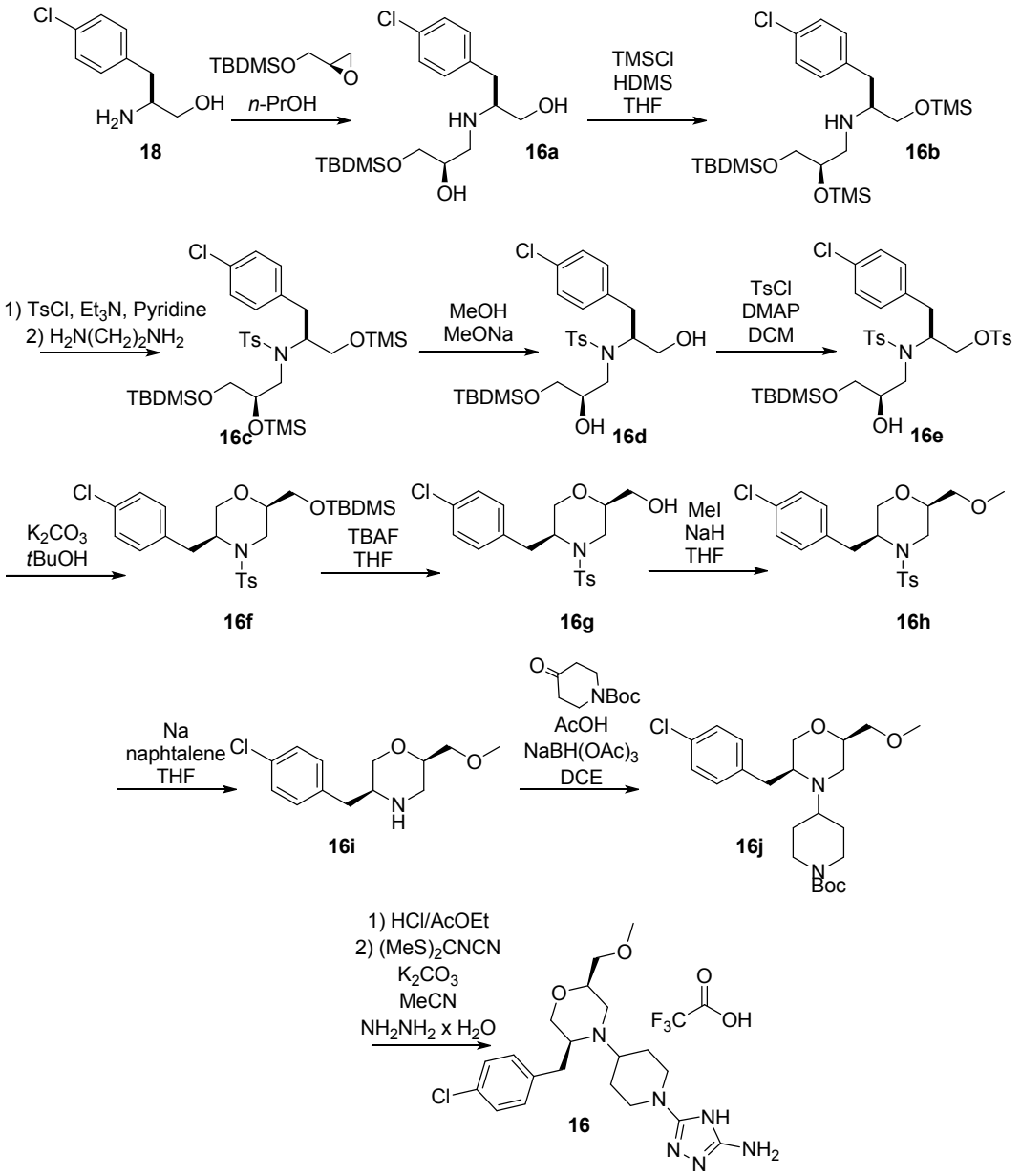
ESI-MS m/z for $C_{24}H_{38}ClN_2O_4$ found 453.1/455.1 $[M+H]^+$; ^{1}H NMR (500 MHz, DMSO-d_6 + D_2O) δ 7.29 ($J_{AA'BB'}$ = 8.3 Hz, 2H), 7.19 ($J_{AA'BB'}$ = 8.1Hz, 2H), 3.92-3.80 (m, 2H), 3.66-3.55 (m, 3H), 3.45-3.39 (m, 1H), 3.35-3.29 (m, 1H), 3.20-3.13 (m, 1H), 2.84-2.75 (m, 3H), 2.72-2.65 (m, 2H), 1.97-1.84 (m, 2H), 1.69-1.61 (m, 2H), 1.33 (s, 9H), 1.09 (s, 3H), 1.07 (s, 3H).

Step 10.

Synthesis of 2-((2*R*,5*S*)-4-(1-(5-amino-4*H*-1,2,4-triazol-3-yl)piperidin-4-yl)-5-(4-chlorobenzyl)morpholin-2-yl)propan-2-ol 2,2,2-trifluoroacetate (**15**).

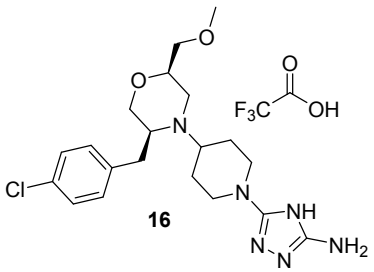
The title compound (**15**) was obtained as a TFA salt according to the General Procedure B from compound **15i** (344 mg; 0.76 mmol) in 30% yield (125 mg; 0.23 mmol).

^{1}H NMR (600 MHz, DMSO-d_6 + D_2O) δ 7.39 ($J_{AA'BB'}$ = 8.5 Hz, 2H), 7.31 ($J_{AA'BB'}$ = 8.5 Hz, 2H), 3.89-3.77 (m, 2H), 3.75-3.69 (m, 1H), 3.67-3.53 (m, 3H), 3.46-3.37 (m, 2H), 3.21-2.97 (m, 3H), 2.97-2.86 (m, 2H), 2.25-2.16 (m, 2H), 1.64-1.55 (m, 2H), 1.15 (s, 3H), 1.14 (s, 3H); ^{1}H NMR (700 MHz, DMSO-d_6 + D_2O, 348 K) δ 7.45 – 7.35 (m, 2H), 7.35 – 7.26 (m, 2H), 3.96 – 3.84 (m, 2H), 3.76 – 3.62 (m, 4H), 3.54 – 3.50 (m, 2H), 3.22 – 3.16 (m, 1H), 3.12 – 3.06 (m, 2H), 2.97 – 2.87 (m, 2H), 2.30 – 2.13 (m, 2H), 1.73 – 1.61 (m, 2H), 1.19 (s, 6H); ^{13}C NMR (176 MHz, DMSO-d_6) δ 134.9, 132.4, 132.0 (2x), 129.2 (2x), 70.4, 65.0, 59.4, 59.01, 57.7, 55.4, 52.1, 45.3, 45.2, 44.0, 40.0, 27.2, 27.1, 25.6, 24.6; HRMS (ESI) *m/z* calc. for $C_{21}H_{32}ClN_6O_2$ $[M+H]^+$ 435.2270 found 435.2282.



Example 16.

Synthesis of 5-(4-((2*R*,5*S*)-5-(4-chlorobenzyl)-2-(methoxymethyl)morpholino)piperidin-1-yl)-4*H*-1,2,4-triazol-3-amine 2,2,2-trifluoroacetate (**16**).



Step 1.

Synthesis of (*S*)-2-(((*R*)-3-((*tert*-butyldimethylsilyl)oxy)-2-hydroxypropyl)amino)-3-(4-chlorophenyl)propan-1-ol (**16a**).

A solution of (*R*)-*tert*-butyldimethyl(oxiran-2-ylmethoxy)silane (5 mL; 26.54 mmol) and (*S*)-2-amino-3-(4-chlorophenyl)propan-1-ol (**18**) (7.4 g; 39.81 mmol) in 1-propanol (100 mL) was heated at reflux for 5 hours and then at room temperature over the weekend. The resulting yellow solution was concentrated *in vacuo* and the crude product was purified by flash column chromatography (AcOEt/MeOH 100:0 to 1:1 *v*/*v*) and **16a** was obtained as a white solid in 68% yield (6.7 g; 17.95 mmol). ESI-MS m/z for $C_{18}H_{33}ClNO_3Si$ found 374.0/376.0 $[M+1]^+$; ^{1}H NMR (700 MHz, $CDCl_3$) δ 7.30 – 7.26 (m, 2H), 7.17 – 7.12 (m, 2H), 4.17 – 4.10 (m, 1H), 3.75 – 3.70 (m, 1H), 3.65 – 3.59 (m, 2H), 3.56 – 3.49 (m, 1H), 3.36 – 3.29 (m, 1H), 2.91 – 2.83 (m, 1H), 2.79 – 2.70 (m, 2H), 2.70 – 2.68 (m, 2H), 2.06 (s, 1H), 1.31 – 1.23 (m, 1H), 0.91 (s, 9H), 0.08 (s, 6H).

Step 2.

Synthesis of (*R*)-3-((*tert*-butyldimethylsilyl)oxy)-*N*-((*S*)-1-(4-chlorophenyl)-3-((trimethylsilyl)oxy)propan-2-yl)-2-((trimethylsilyl)oxy)propan-1-amine (**16b**).

Hexamethyldisilazane (HMDS; 7.6 mL; 36.72 mmol) and trimethylsilyl chloride (TMSCl; 0.45 mL; 3.58 mmol) were added sequentially to a solution of amino diol **16a** (6.7 g; 17.91 mmol) in THF (180 mL) at 0°C. After 2 minutes, the cooling bath was removed, and the resulting white suspension was stirred at room temperature for 70 minutes, then additional portion of TMSCl (0.8 mL; 6.44 mmol) was added and suspension was stirred for further 30 minutes. The reaction mixture was partitioned between ether and a 1/1 mixture of aqueous phosphate buffer solution (0.05 M) and brine (200 mL). The organic layer was separated and the aqueous layer was extracted with ether. The combined organic extracts were dried over $MgSO_4$ and concentrated, providing bis-trimethylsilyl ether **16b** as a light yellow liquid in 99% yield (9.19 g; 17.73 mmol).

^{1}H NMR (700 MHz, $CDCl_3$) δ 7.27 – 7.24 (m, 2H), 7.17 – 7.14 (m, 2H), 3.80 – 3.74 (m, 1H), 3.57 – 3.48 (m, 3H), 3.47 – 3.42 (m, 1H), 2.84 – 2.77 (m, 3H), 2.70 – 2.65 (m, 1H), 2.60 – 2.56 (m, 1H), 1.90 – 1.86 (m, 1H), 0.94 – 0.86 (m, 9H), 0.14 – 0.11 (m, 9H), 0.11 – 0.09 (m, 9H), 0.07 – 0.05 (m, 6H).

Step 3.

Synthesis of *N*-((*R*)-3-((*tert*-butyldimethylsilyl)oxy)-2-((trimethylsilyl)oxy)propyl)-*N*-((*S*)-1-(4-chlorophenyl)-3-((trimethylsilyl)oxy)propan-2-yl)-4-methylbenzenesulfonamide (**16c**).

Triethylamine (15.5 mL; 107.46 mmol) and tosyl chloride (TsCl; 10.2 g; 53.73 mmol) were added sequentially to a solution of bis-trimethylsilyl ether **16b** (9.19 g; 17.73 mmol) in pyridine (89 mL). The resulting red-orange solution was stirred at room temperature for 80 minutes. Ethylene diamine (3.6 mL; 53.73 mmol) was then added and the resulting solution was stirred for 12 hours at room temperature. The solution was then diluted with ethyl acetate, and washed with saturated solution of sodium bicarbonate and brine. The organics extracts were combined and dried over $MgSO_4$, filtered, and the solvent was removed *in vacuo* and the crude product was purified by silica-gel column

chromatography (hexane/AcOEt 10:1 *v*/*v*) and **16c** was obtained as a colorless oil in 92% yield (10.97 g; 16.31 mmol).

^{1}H NMR (500 MHz, $CDCl_3$) δ 7.56 – 7.52 (m, 2H), 7.19 – 7.14 (m, 2H), 7.13 – 7.09 (m, 2H), 7.00 – 6.96 (m, 2H), 4.11 – 4.04 (m, 1H), 3.98 – 3.89 (m, 1H), 3.70 – 3.58 (m, 3H), 3.49 – 3.45 (m, 1H), 3.41 – 3.34 (m, 1H), 3.11 – 3.03 (m, 1H), 3.01 – 2.91 (m, 1H), 2.82 – 2.71 (m, 1H), 2.40 (s, 3H), 0.91 (s, 9H), 0.16 (s, 9H), 0.07 (s, 6H), -0.02 (s, 9H).

Step 4.

Synthesis of *N*-((*R*)-3-((*tert*-butyldimethylsilyl)oxy)-2-hydroxypropyl)-*N*-((*S*)-1-(4-chlorophenyl)-3-hydroxypropan-2-yl)-4-methylbenzenesulfonamide (**16d**).

Sodium methoxide (180 mg; 3.26 mmol) was added in one portion to a solution of sulfonamide **16c** (10.97 g; 16.31 mmol) in methanol (160 mL) at room temperature. The resulting solution was stirred for 40 minutes, and then it was concentrated *in vacuo*. The concentrate was partitioned between ethyl acetate and a 1/1 mixture of saturated aqueous solution of ammonium chloride and brine. The organic layer was separated, and the aqueous layer was extracted with additional portion of ethyl acetate. The combined organics were dried over $MgSO_4$, filtered and concentrated *in vacuo* to provide diol **16d** as a white solid in 99% yield (8.6 g; 16.28 mmol).

ESI-MS m/z for $C_{25}H_{39}ClNO_5SSi$ found 528.3/530.3 $[M+1]^+$; ^{1}H NMR (500 MHz, $CDCl_3$) δ 7.78 – 7.69 (m, 2H), 7.31 – 7.23 (m, 2H), 7.26 – 7.13 (m, 2H), 7.06 – 6.94 (m, 2H), 4.28 – 4.19 (m, 1H), 4.10 – 4.04 (m, 1H), 3.93 – 3.85 (m, 1H), 3.73 – 3.68 (m, 1H), 3.60 – 3.44 (m, 4H), 3.26 – 3.18 (m, 1H), 3.12 – 3.04 (m, 1H), 2.70 – 2.62 (m, 1H), 2.62 – 2.54 (m, 1H), 2.41 (s, 3H), 0.91 (s, 9H), 0.08 (s, 6H).

Step 5.

Synthesis of (*S*)-2-(*N*-((*R*)-3-((*tert*-butyldimethylsilyl)oxy)-2-hydroxypropyl)-4-methylphenylsulfonamido)-3-(4-chlorophenyl)propyl 4-methylbenzenesulfonate (**16e**).

To a solution of diol **16d** (8.6 g; 16.28 mmol), triethylamine (Et_3N; 9.4 mL; 65.24 mmol) in DCM, 4-dimethylaminopyridine (DMAP; 0.79 g; 6.52 mmol) and tosyl chloride (TsCl; 3.2 g; 17.12 mmol) were added sequentially and the resulting solution was stirred at room temperature for 1 hour. The reaction was washed with saturated solution of ammonium chloride and brine. The organics were dried over $MgSO_4$ and concentrated *in vacuo*. The oily residue was purified by flash column chromatography (hexane/AcOEt 8:1 *v*/*v*) and **16e** was obtained as a colorless oil in 55% yield (6.13 g; 8.98 mmol).

ESI-MS m/z for $C_{32}H_{44}ClNO_7S_2SiNa$ found 705.4/707.4 $[M+Na]^+$

Step 6.

Synthesis of (2*R*,5*S*)-2-(((*tert*-butyldimethylsilyl)oxy)methyl)-5-(4-chlorobenzyl)-4-tosylmorpholine (**16f**).

Potassium carbonate (2.5 g; 17.96 mmol) was added to a solution of tosylate **16e** (6.13 g; 8.98 mmol) in *tert*-butyl alcohol (50 mL). The resulting mixture was heated at reflux for 2 hours, and then was

partitioned between ethyl acetate and a mixture 1/1 of sat. aqueous solution of ammonium chloride and brine. The organic layer was separated, and the aqueous layer was extracted with additional portion of ethyl acetate. The combined organics were dried over $MgSO_4$ and dried extracts were concentrated *in vacuo*. The residue was purified by flash column chromatography (hexane/AcOEt 20:1 *v*/*v*) and **16f** was obtained as a white solid in 28% yield (1.3 g; 2.55 mmol).

ESI-MS m/z for $C_{25}H_{36}ClNO_4SSiNa$ found 533.4/535.4 $[M+Na]^+$; ^1H NMR (500 MHz, $CDCl_3$) δ 7.61 – 7.55 (m, 2H), 7.28 – 7.19 (m, 4H), 7.12 – 7.04 (m, 2H), 3.99 – 3.91 (m, 1H), 3.72 – 3.55 (m, 4H), 3.44 – 3.34 (m, 2H), 3.08 (dd, *J* = 13.4, 11.1 Hz, 1H), 2.98 (dd, *J* = 13.3, 9.8 Hz, 1H), 2.76 (dd, *J* = 13.3, 5.6 Hz, 1H), 2.41 (s, 3H), 0.89 (s, 9H), 0.09 – 0.00 (m, 6H).

Step 7.

Synthesis of ((2*R*,5*S*)-5-(4-chlorobenzyl)-4-tosylmorpholin-2-yl)methanol (**16g**).

Solution of *N*-tosyl morpholine **16f** (0.63 g; 1.24 mmol) in THF (2 mL) was treated with tetrabutylammonium fluoride (TBAF) (2.5 mL; 2.46 mmol; 1 M in THF) at room temperature for 2 hours. The reaction mixture was absorbed onto silica gel and purified by column chromatography (AcOEt/hexanes 5:1 *v*/*v*, then AcOEt neat) to give alcohol **16g** in 99% yield (0.49 g; 1.23 mmol).

ESI-MS m/z for $C_{19}H_{23}ClNO_4S$ found 396.0/398.0 $[M+1]^+$

Step 8.

Synthesis of (2*R*,5*S*)-5-(4-chlorobenzyl)-2-(methoxymethyl)-4-tosylmorpholine (**16h**).

To a solution of alcohol **16g** (0.8 g; 2.02 mmol) in dry THF (20 mL), sodium hydride (50% in oil; 145 mg; 3.03 mmol) was added. After 5 minutes MeI (0.18 mL; 3.03 mmol) was added and the reaction mixture was stirred at room temperature for 1 hour. The reaction progress was monitored by TLC and LC-MS. Then another part of NaH (290 mg; 6.06 mmol) and MeI (0.2 mL; 3.37 mmol) were added and the reaction mixture was stirred at room temperature overnight. TLC showed completion of the reaction. The mixture was then poured into a saturated solution of ammonium chloride and extracted 3 times with diethyl ether. Combined organic extracts were washed with brine, dried over anhydrous $MgSO_4$, filtered and concentrated *in vacuo*. The residue was purified by column chromatography (hexane/AcOEt 10:1 to 1:1 *v*/*v*) and **16h** was obtained as a white solid in 95% yield (0.78 g; 1.91 mmol).

ESI-MS m/z for $C_{20}H_{25}ClNO_4S$ found 410.0/412.0 $[M+1]^+$; ^1H NMR (500 MHz, $CDCl_3$) δ 7.57 (d, *J* = 8.3 Hz, 2H,), 7.24 (d, *J* = 8.1 Hz, 2H), 7.20 (d, *J* = 8.4 Hz, 2H), 7.08 (d, *J* = 8.4 Hz, 2H), 3.97-3.91 (m, 1H), 3.70-3.66 (m, 1H), 3.62 (dd, *J* = 2.8, 13.3 Hz, 1H), 3.59-3.54 (m, 1H), 3.48-3.42 (m, 2H), 3.47-3.41 (m, 1H), 3.37 (s, 3H), 3.06 (dd, *J* = 11.1, 13.1 Hz, 1H), 2.97 (dd, *J* = 9.6, 13.3 Hz, 1H), 2.74 (dd, 5.6, 13.3 Hz, 1H), 2.40 (s, 3H).

Step 9.

Synthesis of (2*R*,5*S*)-5-(4-chlorobenzyl)-2-(methoxymethyl)morpholine (**16i**).

Naphthalene (1.22 g; 9.5 mmol) was added in one portion to a vigorously stirred suspension of sodium (0.27 g; 11.89 mmol) in dry THF (5.9 mL; 0.5 mL/1 mmol Na). The resulting green suspension was stirred for 2 hours at room temperature. Then the green solution was added dropwise into solution of amide **16h** (0.78 g; 1.91 mmol) in THF (28 mL) at -70°C until reaction solution changed to dark-green (2 mL of $NaC_{10}H_8$ was added). The reaction was quenched after 10 minutes at -70°C with sat. solution of ammonium chloride and allowed to warm to room temperature. Then the mixture was partitioned between ether and mixture of $NaHCO_3$ and brine. The organic phase was separated, dried over $MgSO_4$ and concentrated *in vacuo*. The residue was purified by column chromatography (hexane/AcOEt 2:1, then AcOEt 100:0 and then AcOEt/MeOH 5:1 *v*/*v*) and **16i** was obtained in 70% yield (0.34 g; 1.33 mmol).

ESI-MS m/z for $C_{13}H_{19}ClNO_2$ found 255.8/257.8 $[M+1]^+$; ^{1}H NMR (500 MHz, $CDCl_3$) δ 7.27 ($J_{AA'BB'}$ = 8.3 Hz, 2H), 7.14 ($J_{AA'BB'}$ = 8.3Hz, 2H), 3.83-3.70 (m, 3H), 3.56 (dd, *J* = 6.4, 10.1 Hz, 1H), 3.45 (dd, *J* = 3.9, 10.1 Hz, 1H), 3.39 (s, 3H), 3.09-3.03 (m, 1H), 3.02-2.95 (m, 2H), 2.90 (dd, *J* =7.1, 13.3 Hz, 1H), 2.81 (dd, *J* = 3.0, 12.2 Hz, 1H).

Step 10.

Synthesis of *tert*-butyl 4-((2*R*,5*S*)-5-(4-chlorobenzyl)-2-(methoxymethyl)morpholino)piperidine-1-carboxylate (**16j**).

The title compound (**16j**) was obtained according to the General Procedure A from compound **16i** (0.34 g; 1.33 mmol) in 39% yield (230 mg; 0.52 mmol).

ESI-MS m/z for $C_{23}H_{36}ClN_2O_4$ found 439.2/441.2 $[M+H]^+$

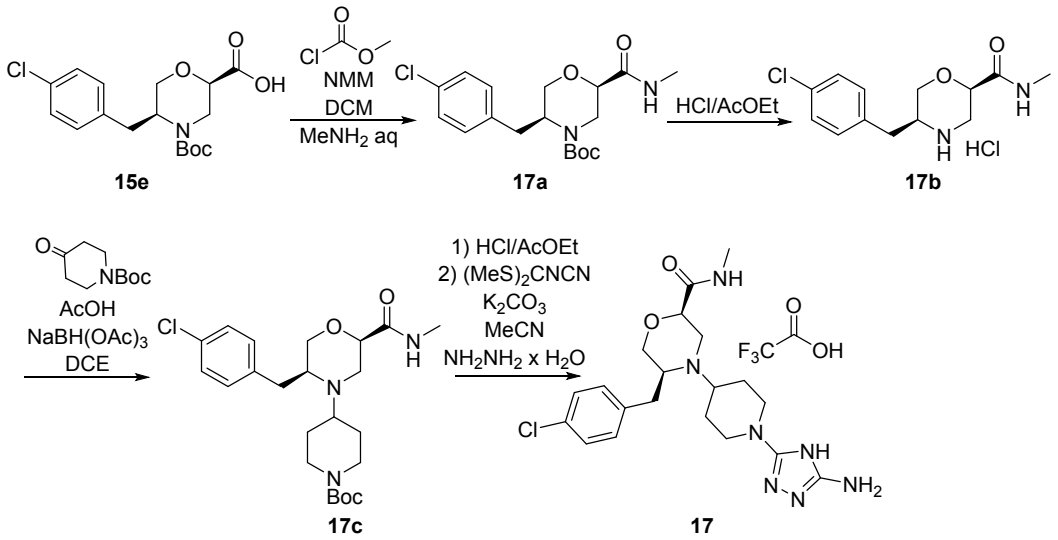
Step 11.

Synthesis of 5-(4-((2*R*,5*S*)-5-(4-chlorobenzyl)-2-(methoxymethyl)morpholino)piperidin-1-yl)-4*H*-1,2,4-triazol-3-amine 2,2,2-trifluoroacetate (**16**).

The title compound (**16**) was obtained as a TFA salt according to the General Procedure B from compound **16j** (408 mg; 0.64 mmol) in 99% yield (335 mg; 0.63 mmol).

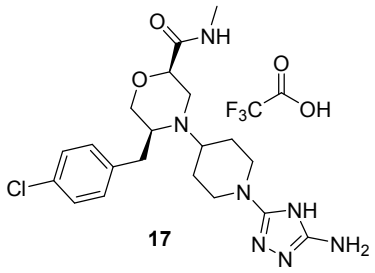
^{1}H NMR (500 MHz, DMSO-d_6) δ 7.42 ($J_{AA'BB'}$ = 8.3Hz, 2H), 7.33 ($J_{AA'BB'}$ = 8.2 Hz, 2H),3.91-3.80 (m, 4H), 3.64-3.53 (m, 3H), 3.53-3.44 (m, 3H), 3.30 (s, 3H), 3.15-3.02 (m, 3H), 2.94-2.86 (m, 1H), 2.85-2.79 (m, 1H), 2.21-2.12 (m, 2H), 1.66-1.55 (m, 2H); ^{1}H NMR (700 MHz, DMSO-d_6 + D_2O, 348 K) δ 7.40 – 7.35 (m, 2H), 7.35 – 7.26 (m, 2H), 3.92 – 3.83 (m, 3H), 3.74 – 3.66 (m, 2H), 3.66 – 3.60 (m, 2H), 3.55 – 3.50 (m, 2H), 3.44 – 3.38 (m, 1H), 3.36 – 3.28 (m, 3H), 3.20 – 3.07 (m, 3H), 2.96 – 2.85 (m, 2H), 2.21 – 2.09 (m, 2H), 1.69 – 1.57 (m, 2H); ^{13}C NMR (176 MHz, DMSO-d_6) δ 134.9, 132.4, 131.9 (2×), 129.2 (2×), 72.2, 64.9, 59.2, 58.84, 58.76, 55.6, 45.2, 45.1, 39.9, 27.4, 25.91, 25.89, 25.6 (one signal is missed due to

overlap); HRMS (ESI) *m/z* calc. for $C_{20}H_{30}ClN_6O_2$ $[M+H]^+$ 421.2113 found 421.2131.



Example 17.

Synthesis of (2*R*,5*S*)-4-(1-(5-amino-4*H*-1,2,4-triazol-3-yl)piperidin-4-yl)-5-(4-chlorobenzyl)-*N*-methylmorpholine-2-carboxamide 2,2,2-trifluoroacetate (**17**).



Step 1.

Synthesis of *tert*-butyl (2*R*,5*S*)-5-(4-chlorobenzyl)-2-(methylcarbamoyl)morpholine-4-carboxylate (**17a**).

Carboxylic acid **15e** (0.37 g; 1.03 mmol) was dissolved in dichloromethane (DCM) (5 ml) and *N*-methylmorpholine (136 mg; 1.24 mmol) was added. The solution was cooled to –15ºC and methyl chloroformate (0.1 mL; 1.24 mmol) was added and the mixture was stirred for additional 10 minutes at which time a solution of methylamine (2 M in THF; 2 mL; 4.12 mmol) was added. The reaction mixture was allowed to warm to room temperature and was stirred overnight. The organic phase was washed subsequentially with 1 M HCl, 1 M NaOH, and brine. The organic phase was dried over $MgSO_4$, filtered and concentrated *in vacuo*. The residue was purified by flash column chromatography (hexane/AcOEt 5:1 *v*/*v*) and **17a** was obtained in 26% yield (0.1 g; 0.27 mmol).

ESI-MS m/z for $C_{18}H_{25}ClN_2O_4Na$ found 391.7/393.7 $[M+Na]^+$

Step 2.

Synthesis of (2*R*,5*S*)-5-(4-chlorobenzyl)-*N*-methylmorpholine-2-carboxamide hydrochloride (**17b**).

Boc-protected morpholine **17a** (0.33 g; 0.89 mmol) was treated with 4M HCl in EtOAc solution (3 mL) and reaction was stirred in room temperature for 1 hour. Volatiles were removed *in vacuo* and **17b** was obtained as a hydrochloride salt in 99% yield (268 mg; 0.88 mmol).

ESI-MS m/z for $C_{13}H_{18}ClN_2O_2$ found 269.2/271.2 $[M+H]^+$

Step 3.

Synthesis of *tert*-butyl 4-((2*R*,5*S*)-5-(4-chlorobenzyl)-2-(methylcarbamoyl)morpholino)piperidine-1-carboxylate (**17c**).

The title compound (**17c**) was obtained according to the General Procedure A from compound **17b** (268 mg; 0.88 mmol) in 63% yield (250 mg; 0.55 mmol).

ESI-MS m/z for $C_{23}H_{35}ClN_3O_4$ found 452.2/454.2 $[M+H]^+$

Step 4.

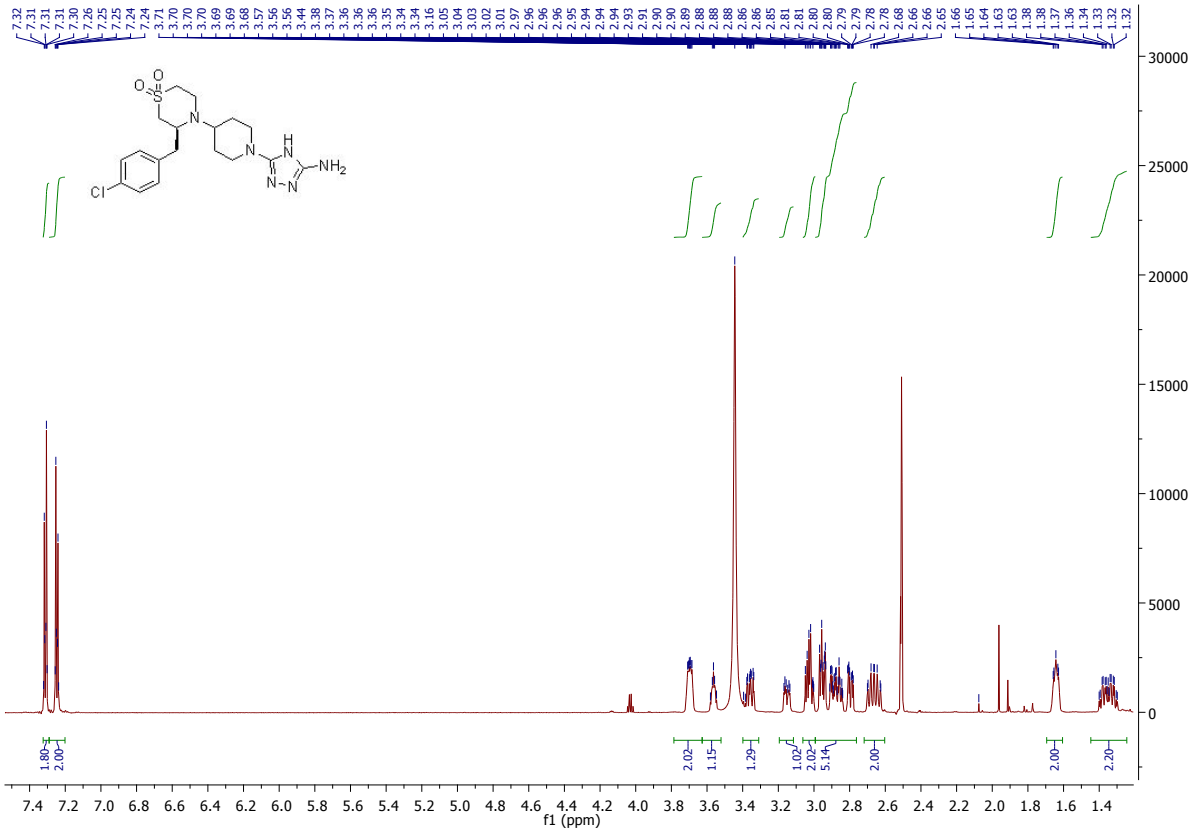
Synthesis of (2*R*,5*S*)-4-(1-(5-amino-4*H*-1,2,4-triazol-3-yl)piperidin-4-yl)-5-(4-chlorobenzyl)-*N*-methylmorpholine-2-carboxamide 2,2,2-trifluoroacetate (**17**).

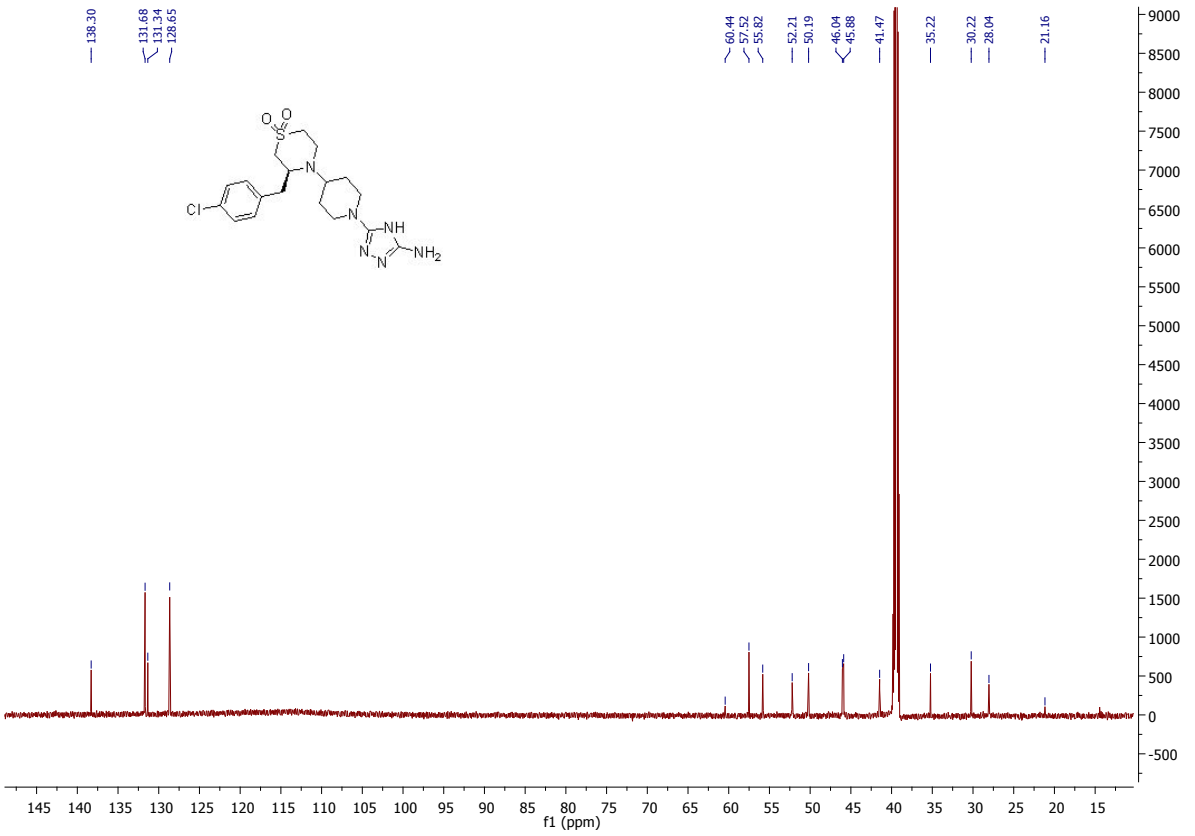
The title compound (**17**) was obtained as a TFA salt according to the General Procedure B from compound **17c** (250 mg; 0.55 mmol) in 53% yield (160 mg; 0.29 mmol).

^{1}H NMR (500 MHz, DMSO-d_6, 348 K) δ 7.62 (bs, 1H), 7.35 ($J_{AA'BB'}$ = 8.5 Hz, 2H), 7.30 ($J_{AA'BB'}$ = 8.5 Hz, 2H), 4.05-3.97 (m, 1H), 3.82-3.74 (m, 2H), 3.63-3.54 (m, 2H), 3.30-3.11 (m, 2H), 3.02-2.82 (m, 6H), 2.66 (d, *J* = 4.7 Hz, 3H), 1.98-1.92 (m, 2H), 1.57-1.47 (m, 2H); ^{1}H NMR (700 MHz, DMSO-d_6 + D_2O, 348 K) δ 7.42 – 7.32 (m, 4H), 4.37 – 4.29 (m, 1H), 3.91 – 3.79 (m, 3H), 3.76 – 3.63 (m, 3H), 3.33 – 3.23 (m, 1H), 3.15 – 3.08 (m, 2H), 3.04 – 2.93 (m, 2H), 2.54 – 2.45 (m, 3H), 2.23 – 2.11 (m, 2H), 1.84 – 1.66 (m, 2H); ^{13}C NMR (176 MHz, DMSO-d_6) δ 167.8, 134.9, 132.4, 132.0 (2×), 129.1 (2×), 73.2, 64.6, 58.7, 55.9, 45.2, 45.1, 44.7, 39.8, 27.9, 26.0, 25.9, 25.5 (one signal is missed due to overlap); HRMS (ESI) *m/z* calc. for $C_{20}H_{29}ClN_7O_2$ $[M+H]^+$ 434.2066 found 434.2071.

^{1}H NMR spectra of compounds 5 - 17.

Example 5.





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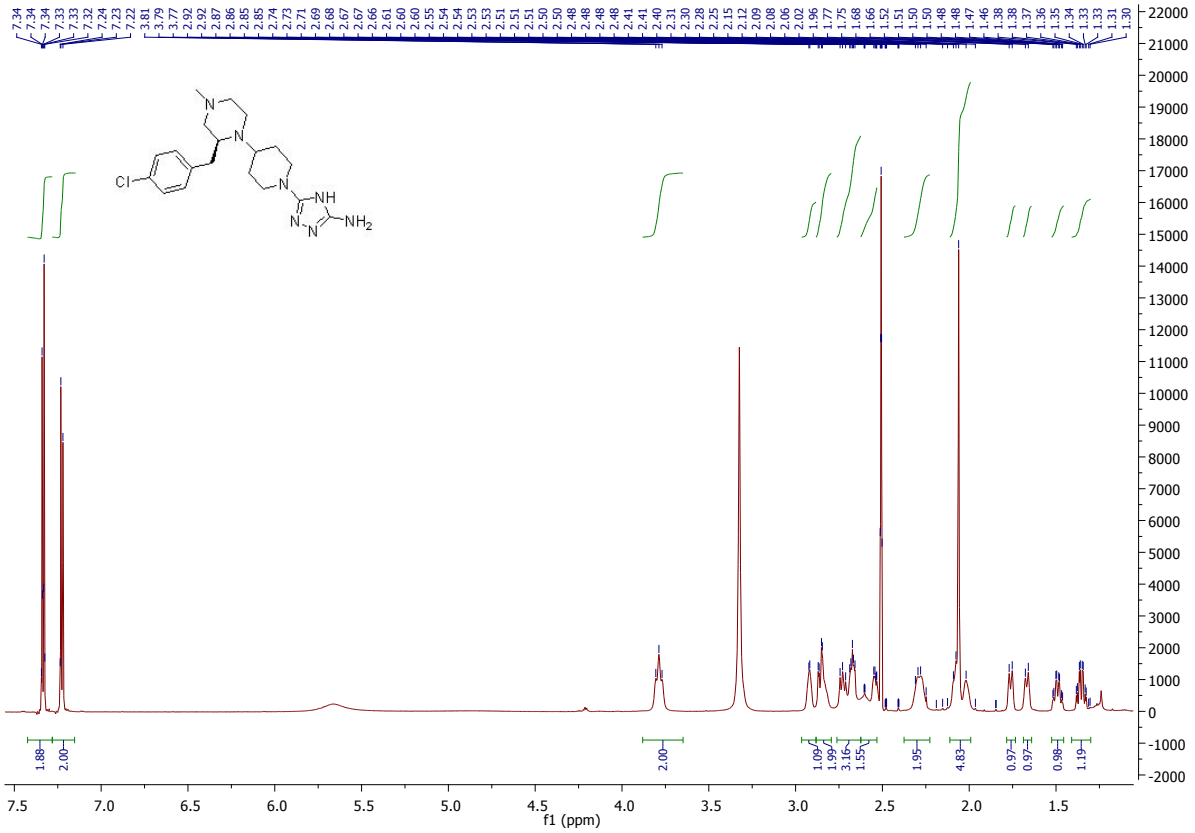

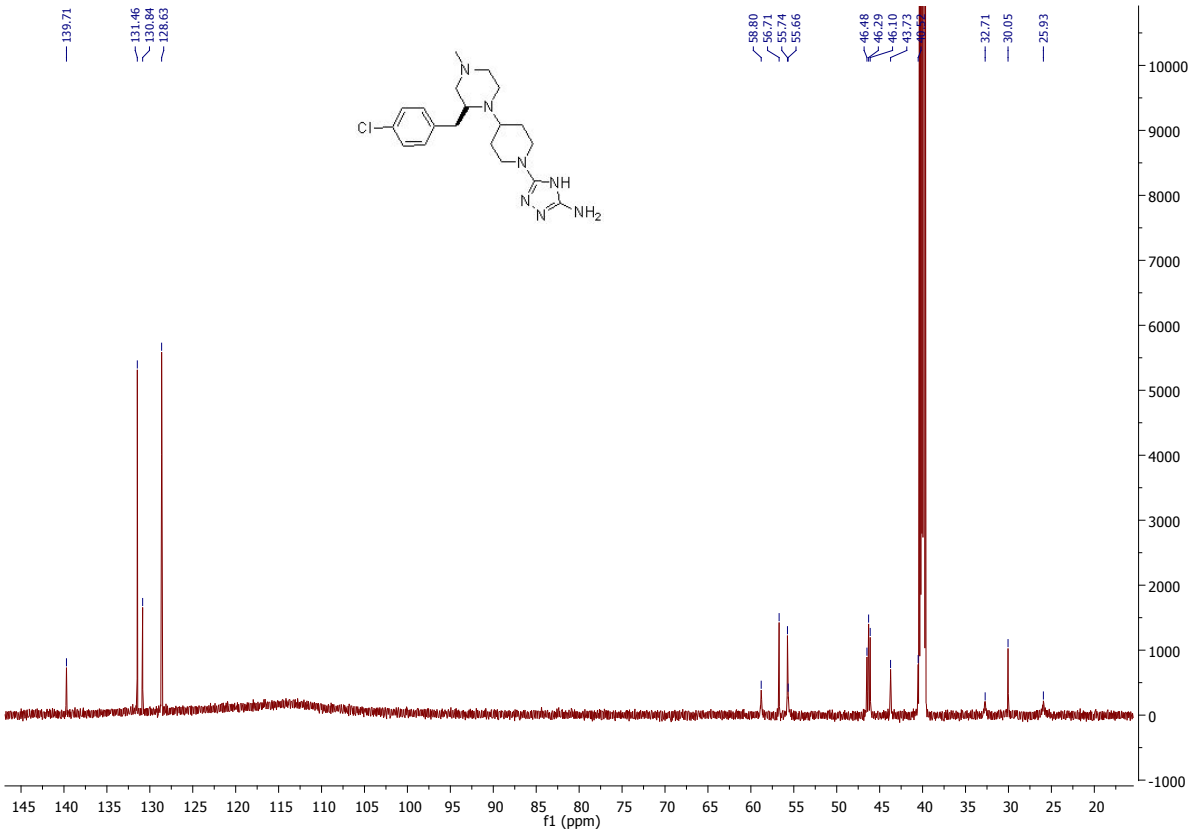

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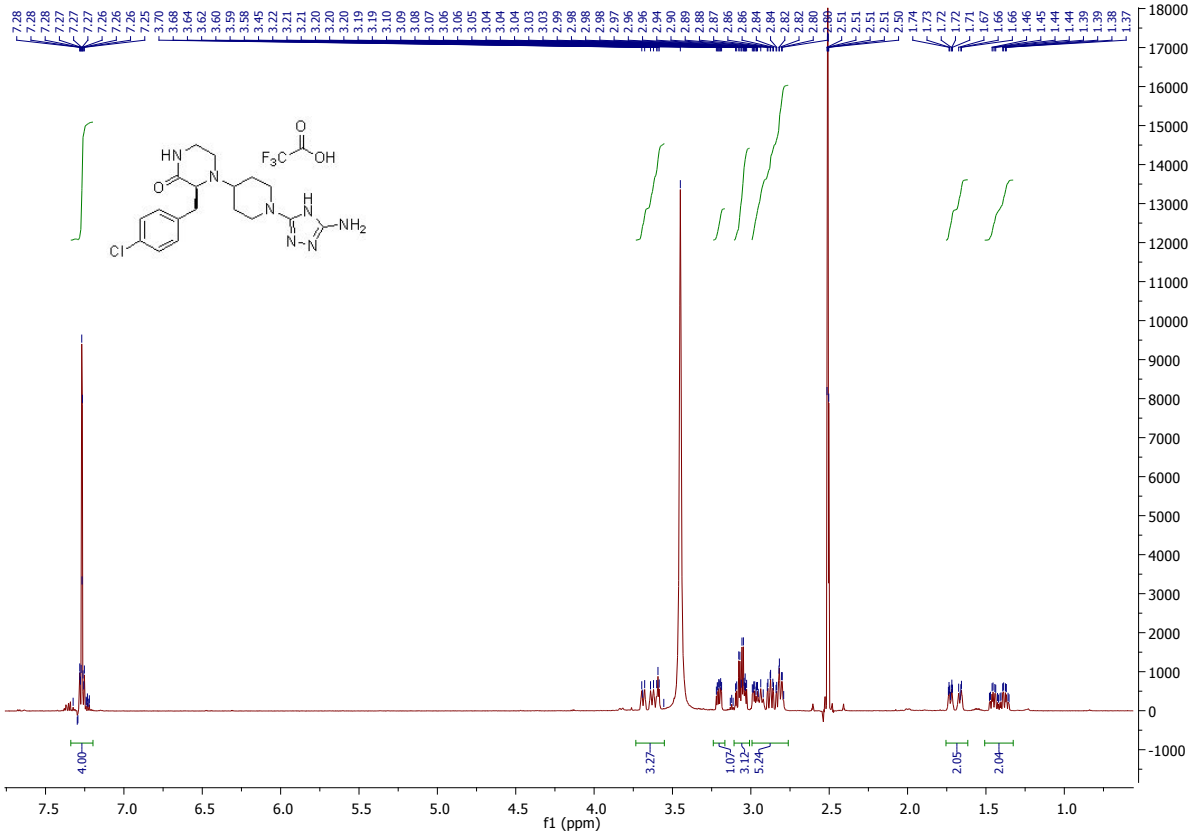
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N
F3C
OH
N
H
N
N
NH2
Cl
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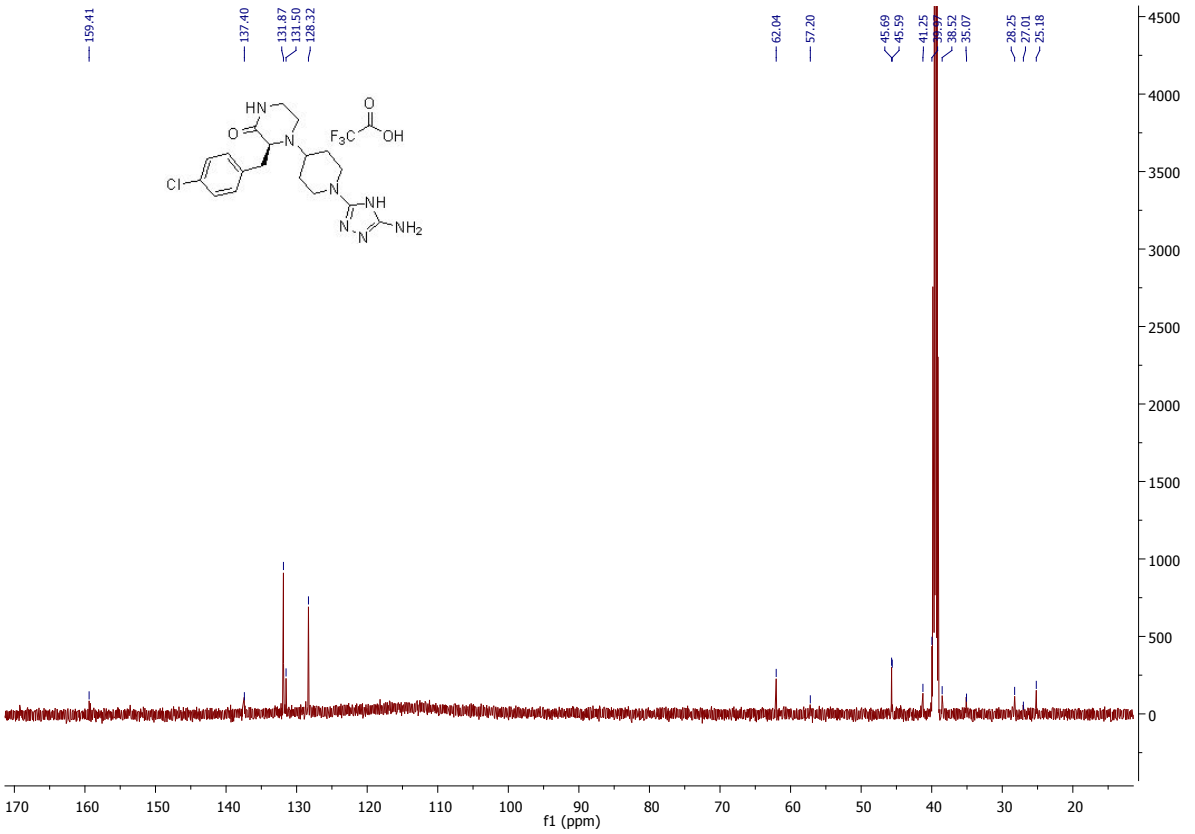
1.07
3.12
5.24
2.05
2.04
f1 (ppm)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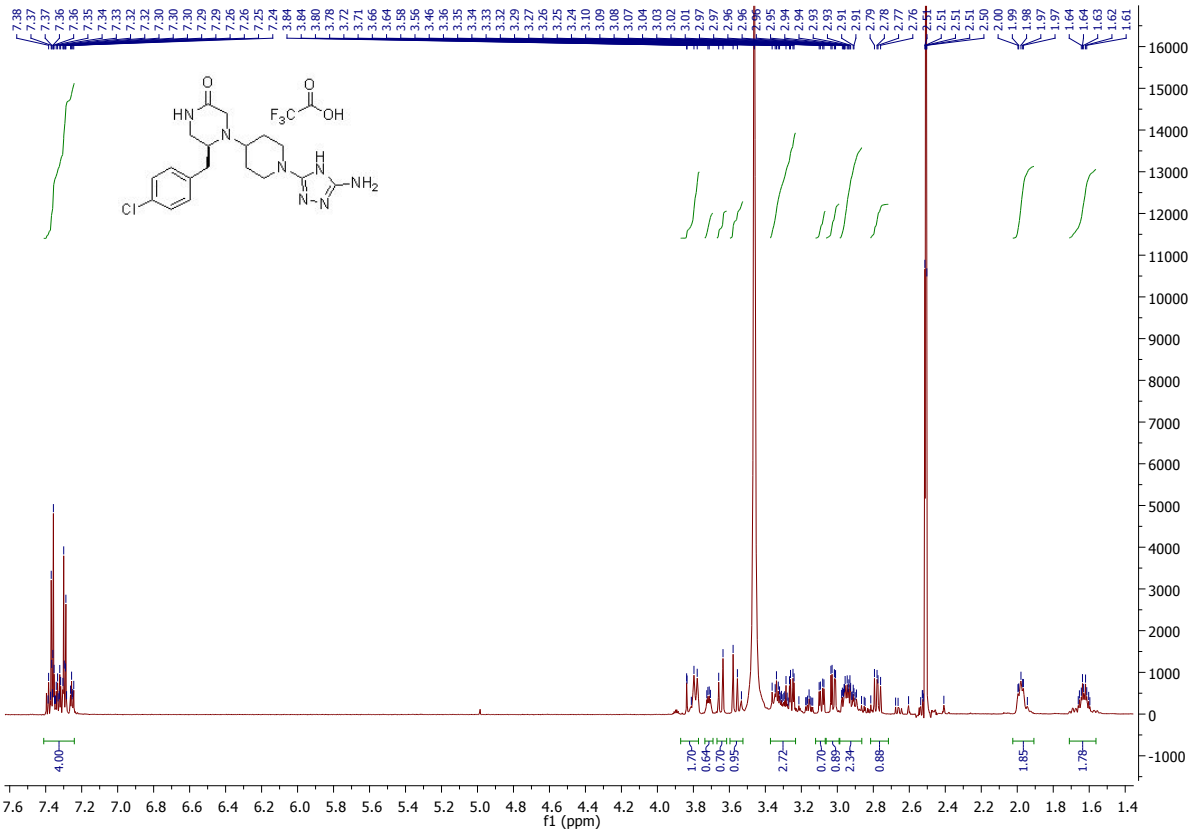
OH
Cl
NH
N
N
NH2
f1 (ppm)

Example 8.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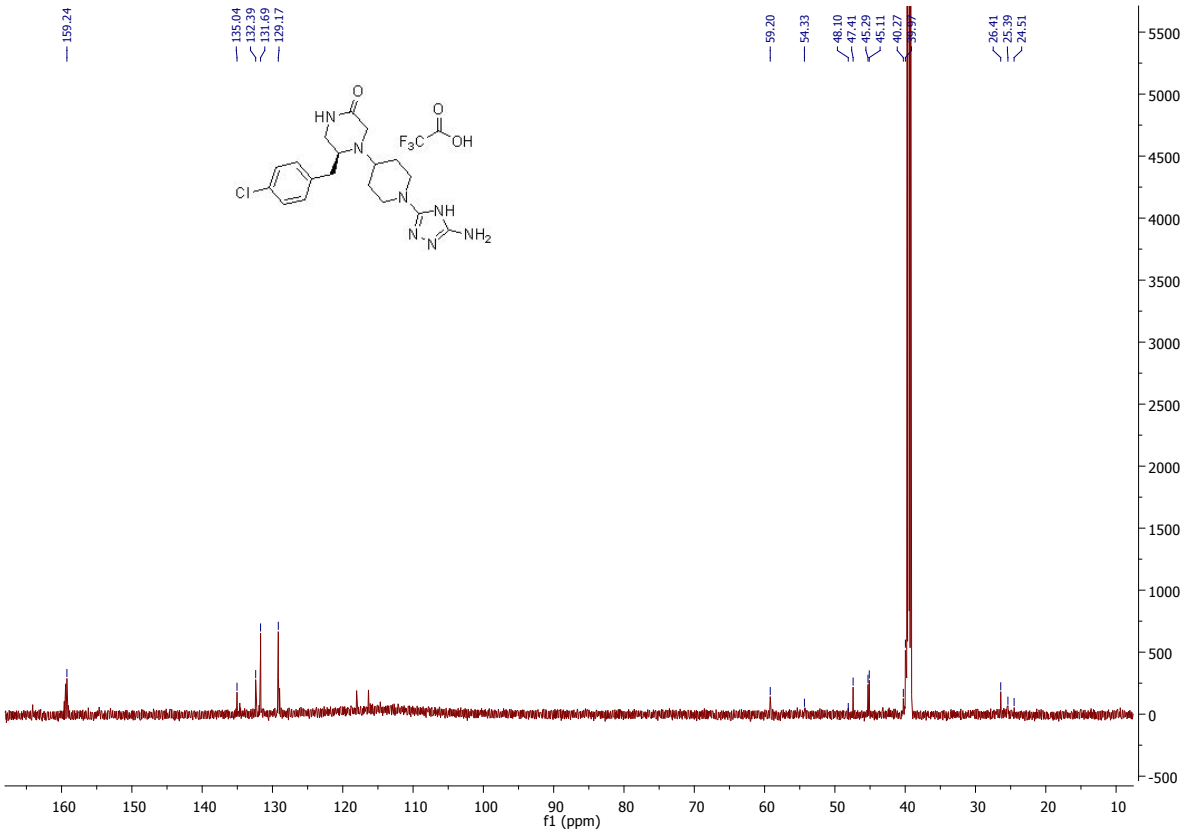
O
F3C
OH
Cl
NH2
f1 (ppm)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47.41
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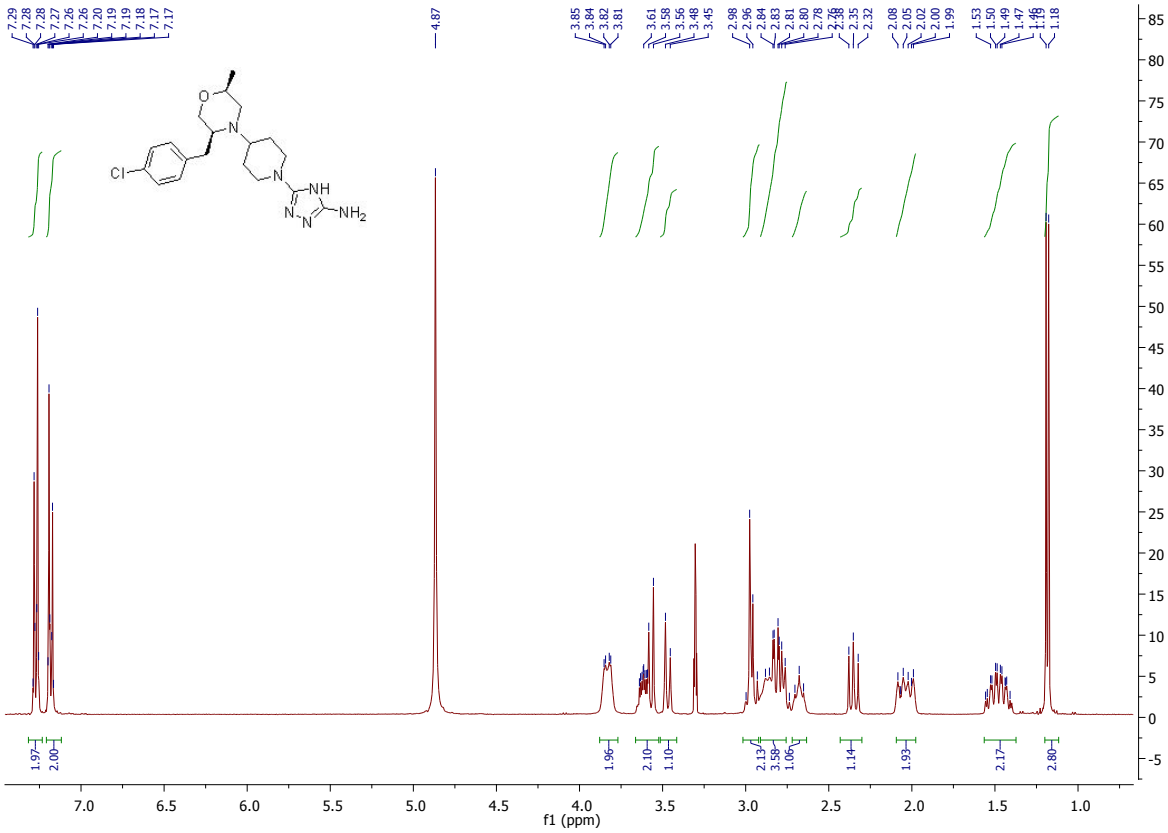
O
F3C
OH
Cl
NH2
f1 (ppm)

Example 9.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7.17
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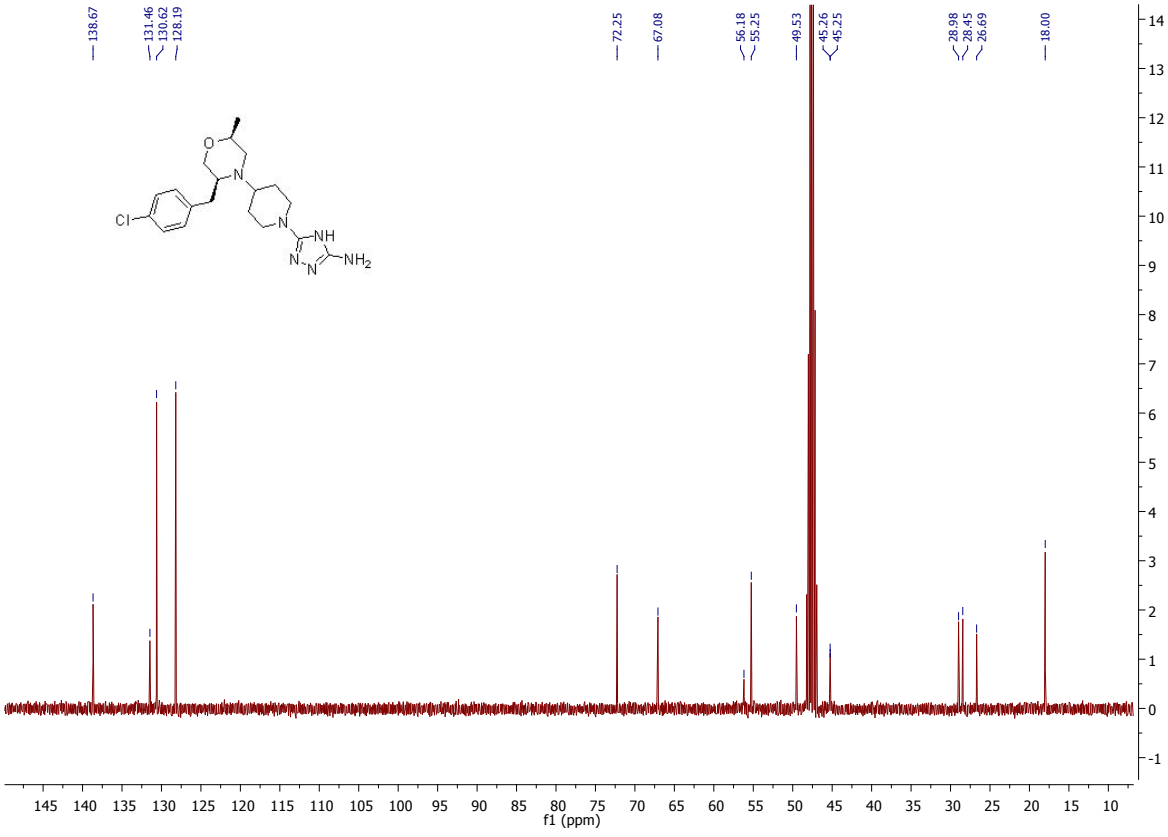
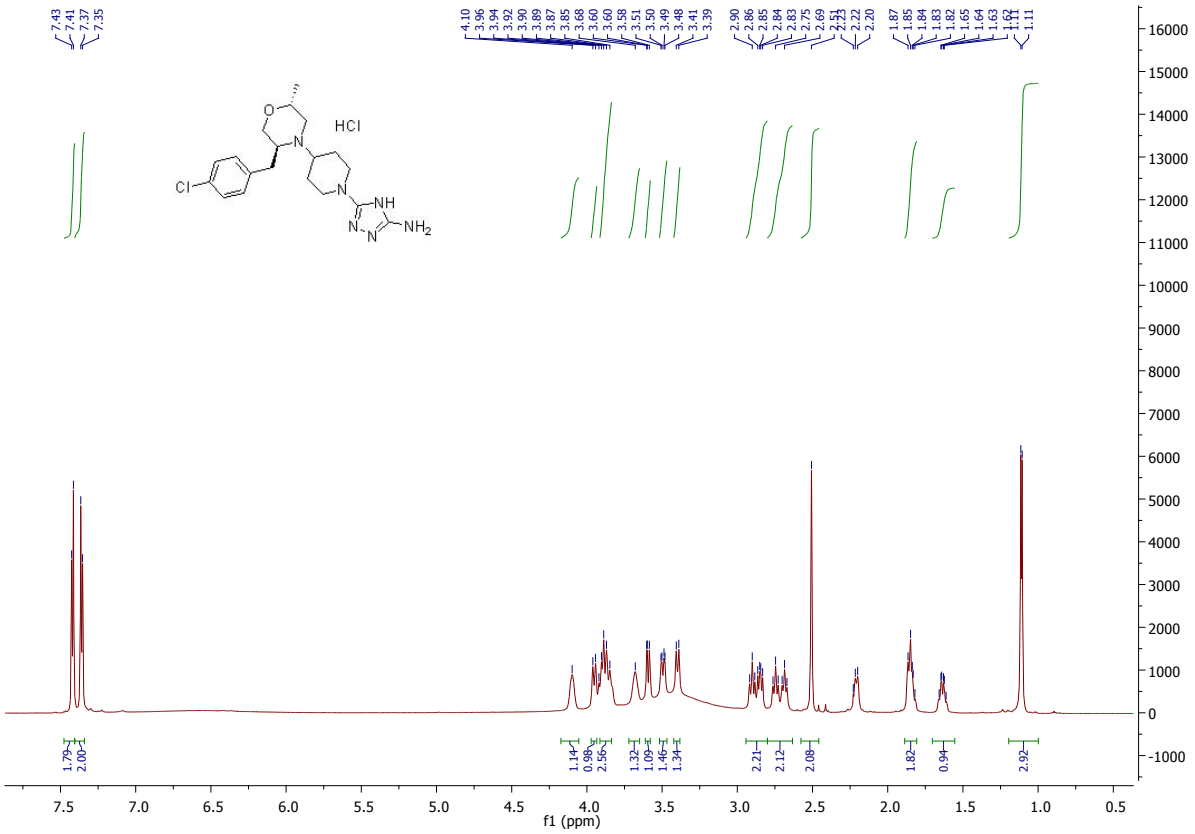
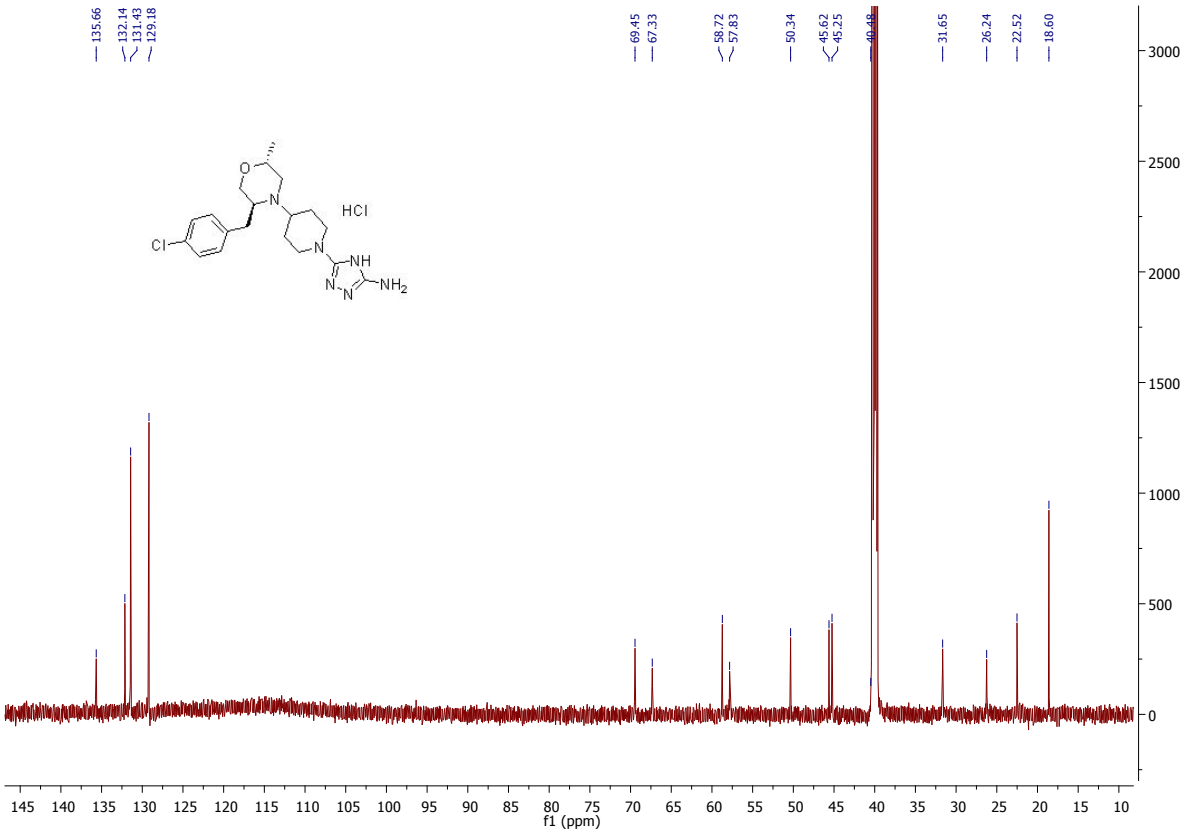
3.81
3.61
3.58
3.56
3.48
3.45
2.98
2.96
2.84
2.83
2.81
2.80
2.78
2.35
2.32
2.08
2.05
2.02
2.00
1.99
1.53
1.50
1.49
1.47
1.18
Cl
O
N
NH
NH2
1.97
2.00
1.96
2.10
1.10
2.13
3.58
1.06
1.14
1.93
2.17
2.80
f1 (ppm)

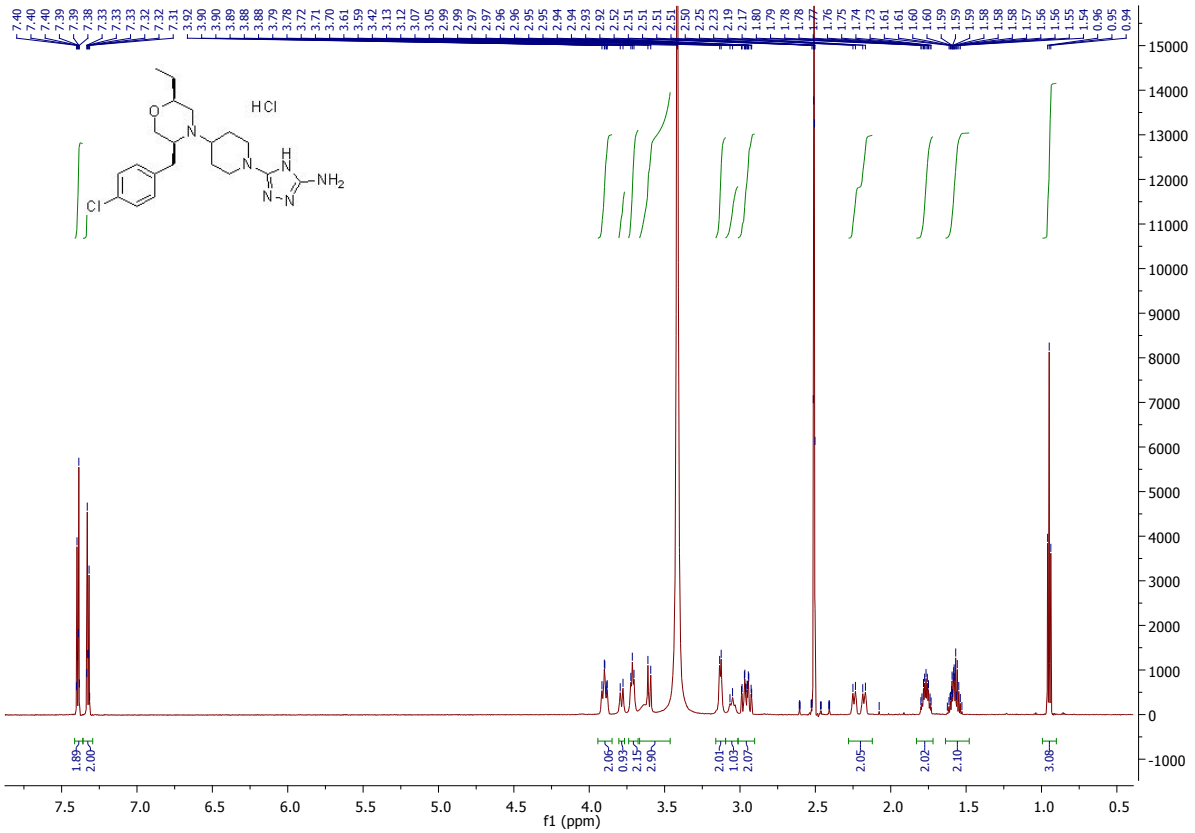

138.67
131.46
130.62
128.19
72.25
67.08
56.18
55.25
49.53
45.26
45.25
28.98
28.45
26.69
18.00
Cl
O
N
NH
NH2
f1 (ppm)

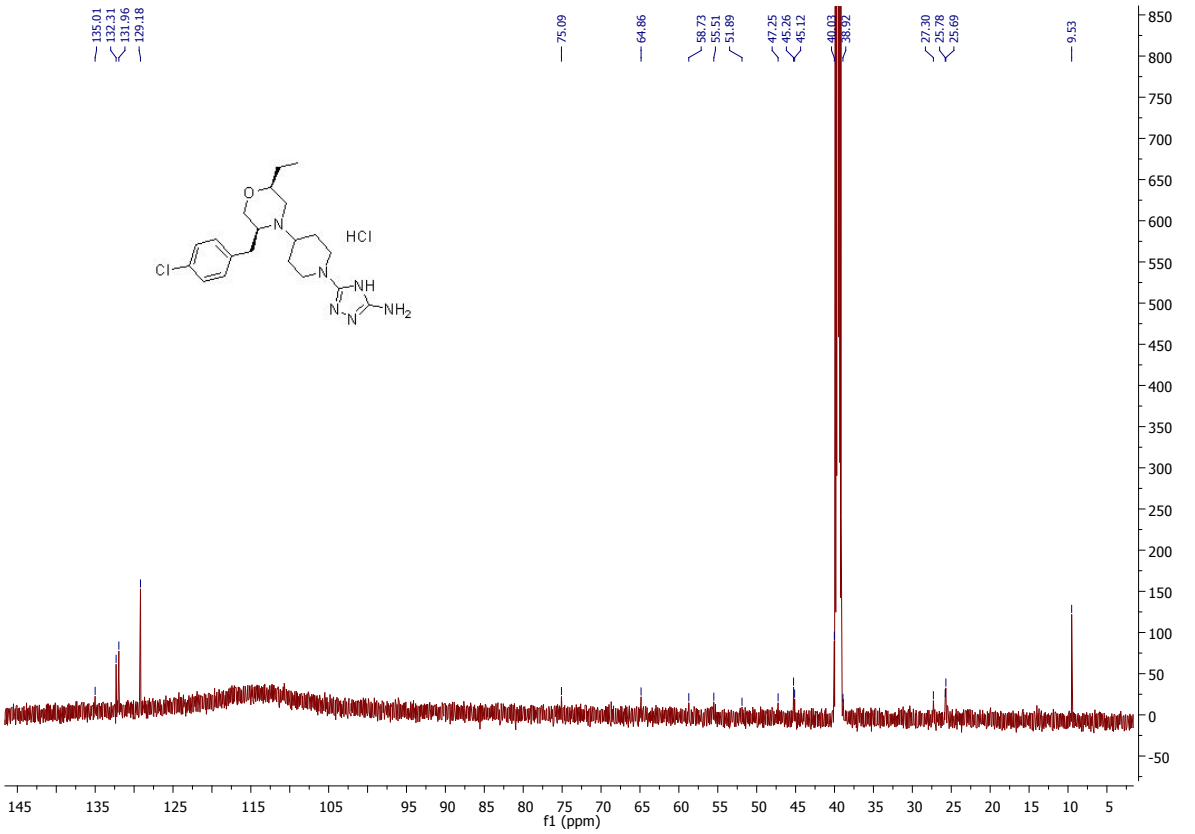
Example 10.


7.43
7.41
7.37
7.35
4.10
3.96
3.94
3.92
3.90
3.89
3.87
3.85
3.68
3.60
3.60
3.58
3.51
3.50
3.49
3.48
3.41
3.39
2.90
2.86
2.85
2.84
2.83
2.75
2.69
2.51
2.23
2.22
2.20
1.87
1.85
1.84
1.83
1.82
1.65
1.64
1.63
1.62
1.11
HCl
Cl
NH
NH2
1.79
2.00
1.14
0.98
2.56
1.32
1.09
1.46
1.34
2.21
2.12
2.08
1.82
0.94
2.92
f1 (ppm)

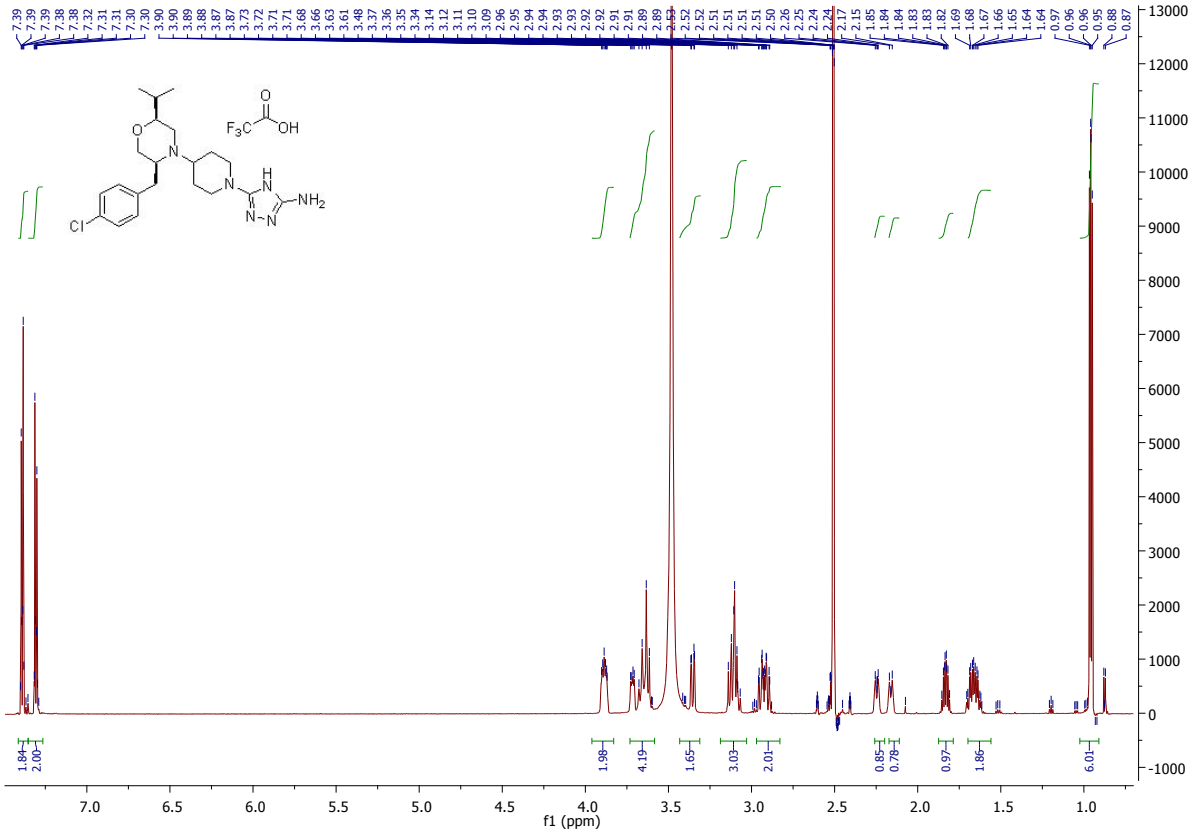

135.66
132.14
131.43
129.18
69.45
67.33
58.72
57.83
50.34
45.62
45.25
40.49
31.65
26.24
22.52
18.60
HCl
Cl
NH
NH2
f1 (ppm)

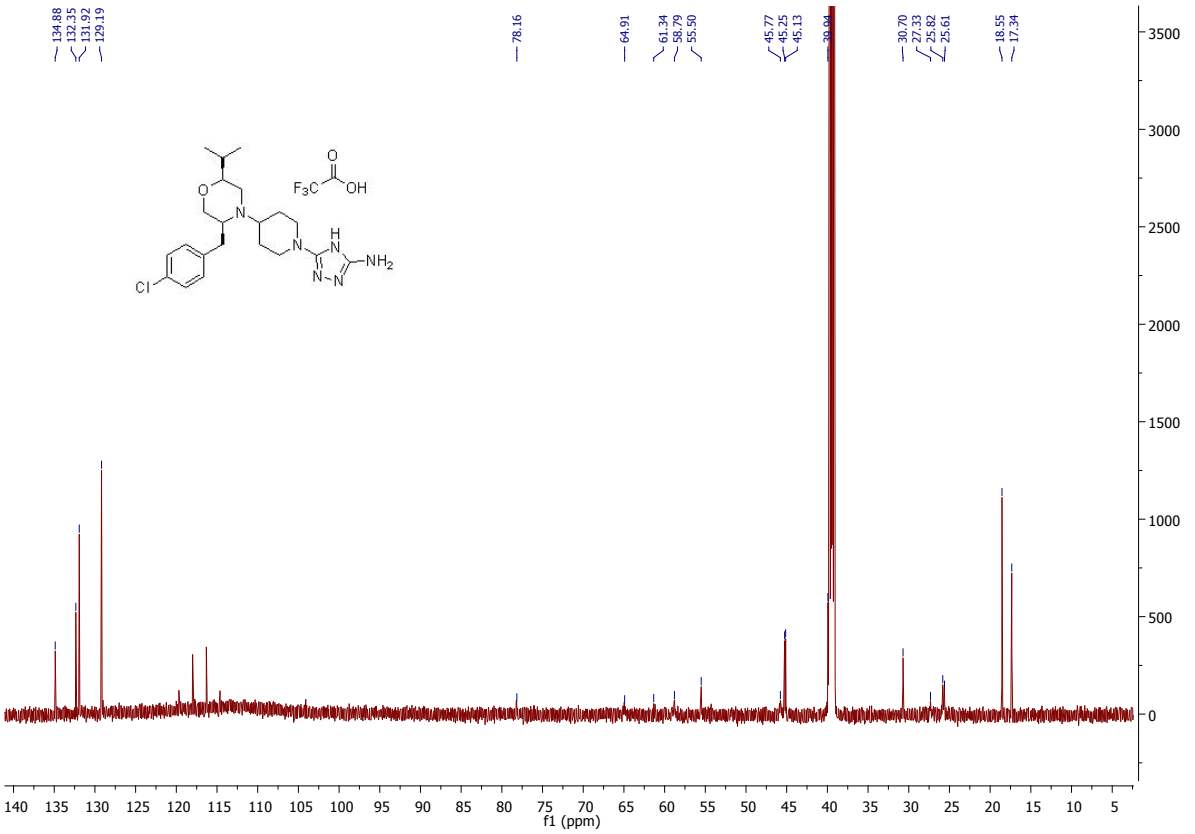
Example 11.



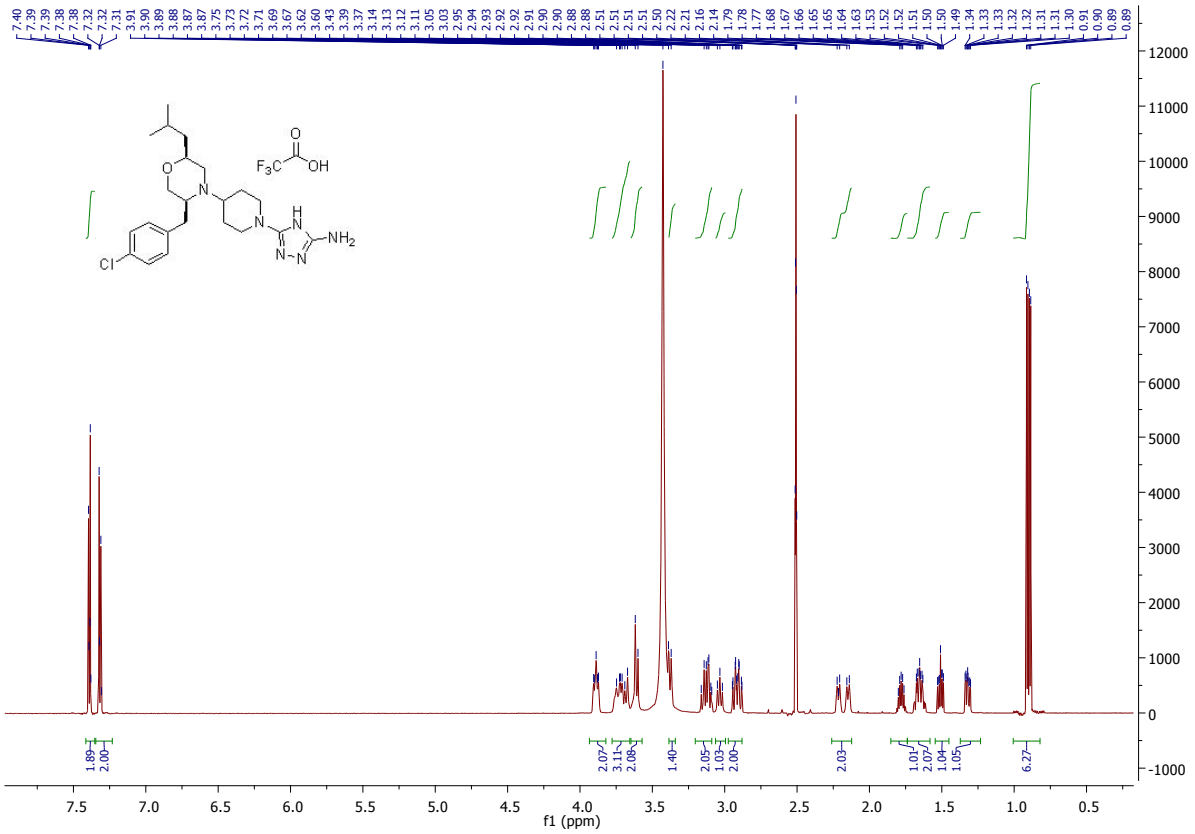


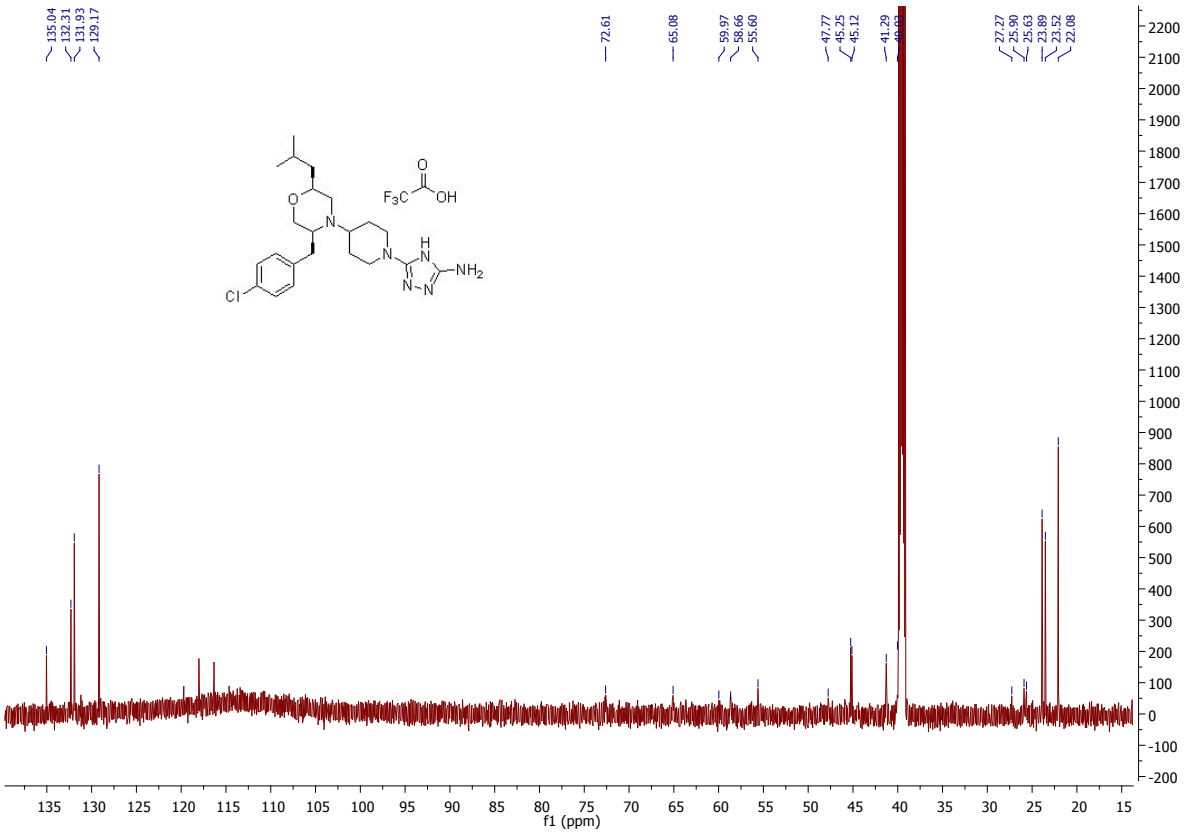
Example 12.



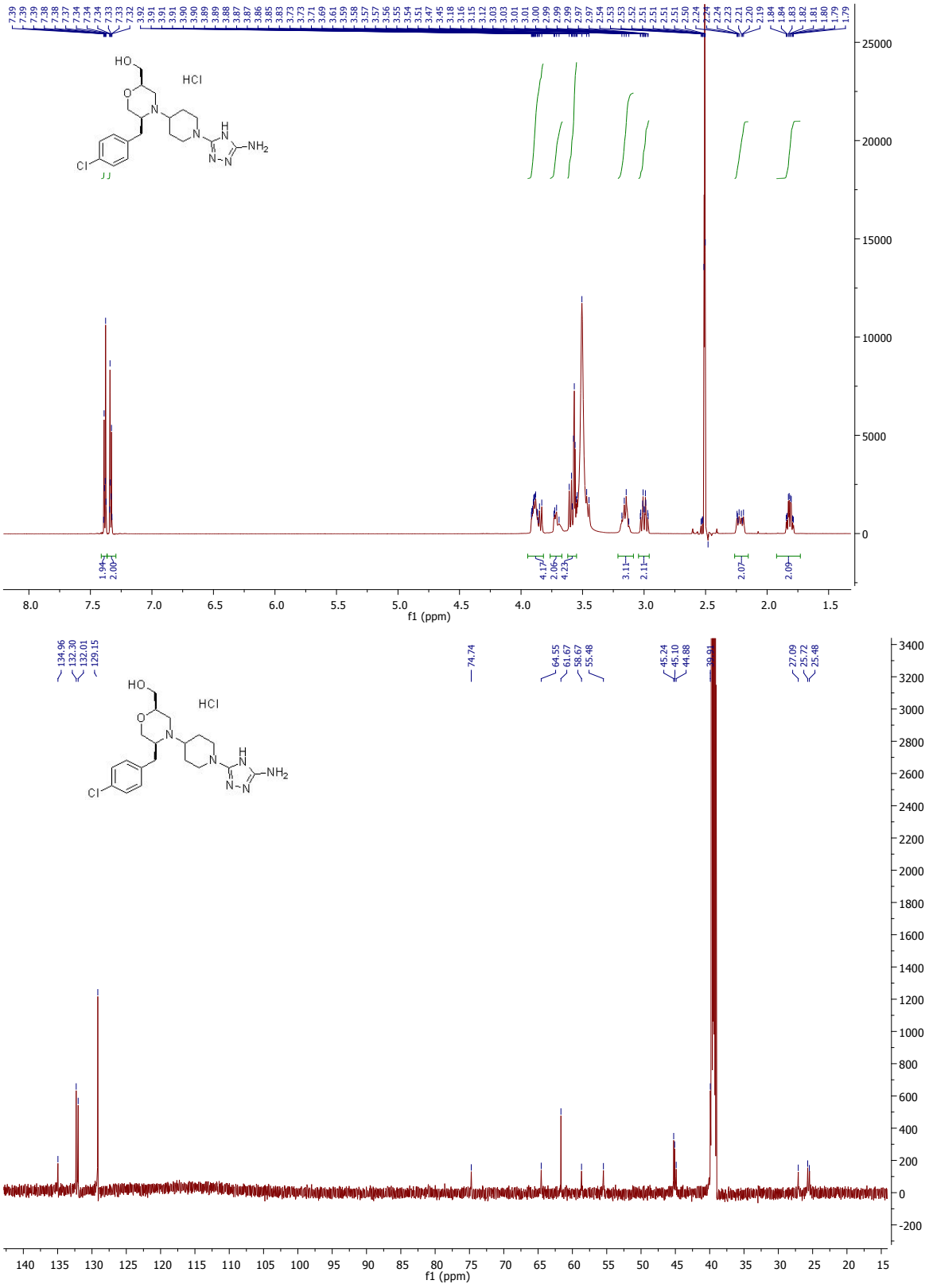


Example 13.

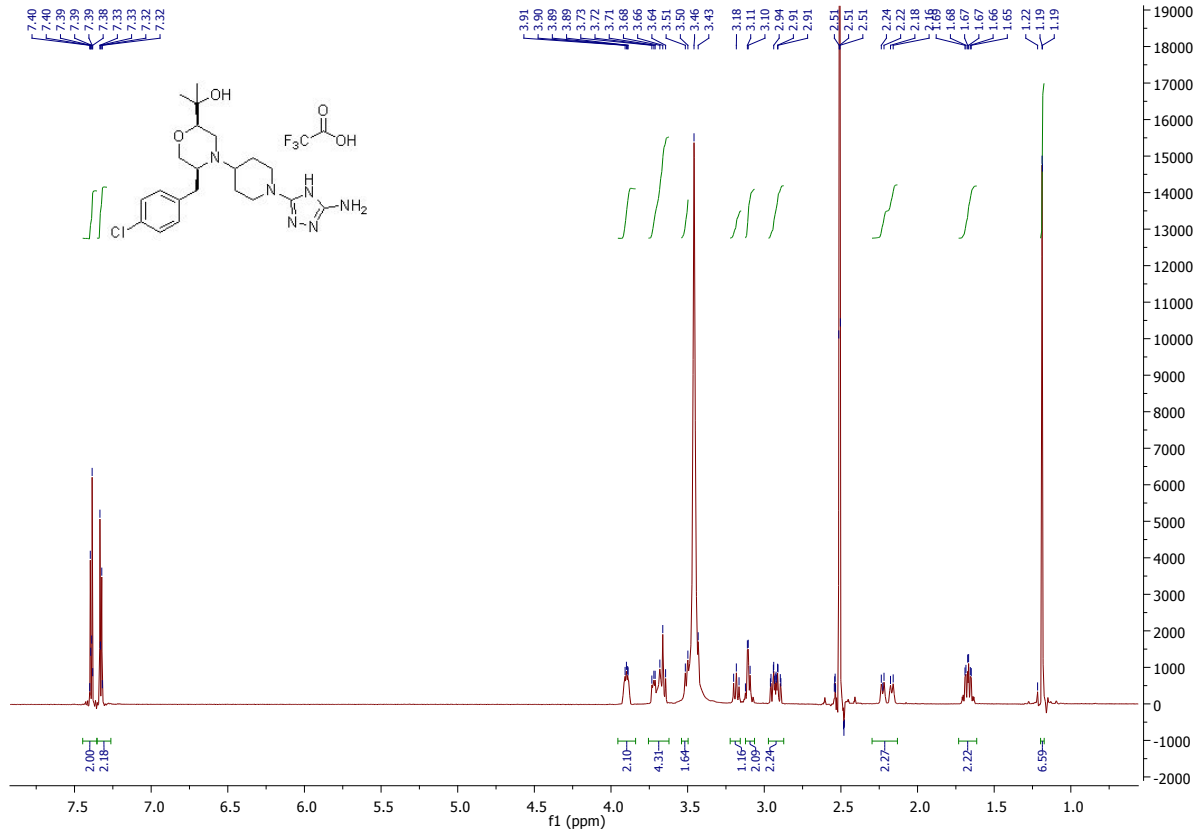




Example 14.



Example 15.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7.39
7.39
7.39
7.38
7.33
7.33
7.32
7.32
3.91
3.90
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3.89
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3.72
3.71
3.68
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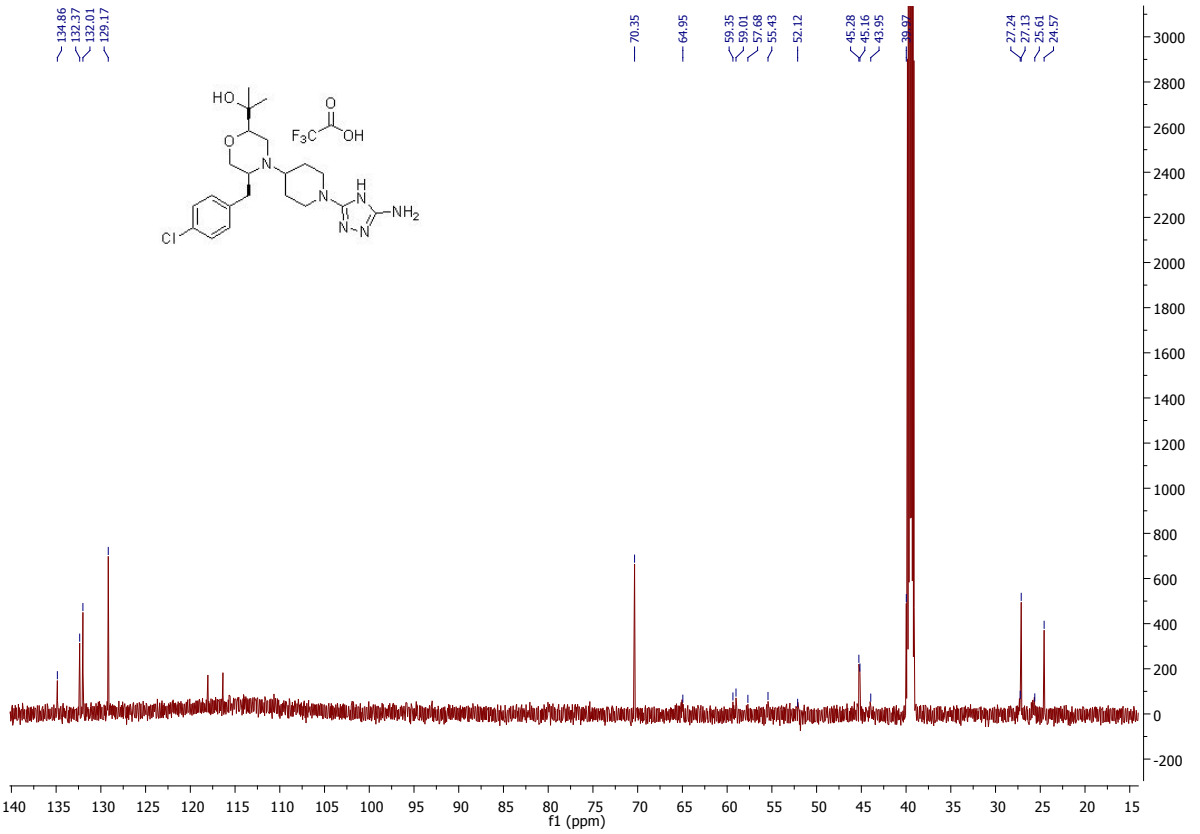
3.64
3.51
3.50
3.46
3.43
3.18
3.11
3.10
2.94
2.91
2.91
2.51
2.51
2.51
2.24
2.22
2.18
2.16
2.16
1.68
1.67
1.67
1.66
1.65
1.22
1.19
1.19
OH
O
F3C
OH
N
N
H
N
N
NH2
Cl
2.00
2.18
2.10
4.31
1.64
1.16
2.09
2.24
2.27
2.22
6.59
f1 (ppm)


134.86
132.37
132.01
129.17
70.35
64.95
59.35
59.01
57.68
55.43
52.12
45.28
45.16
43.95
39.97
27.24
27.13
25.61
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HO
O
F3C
OH
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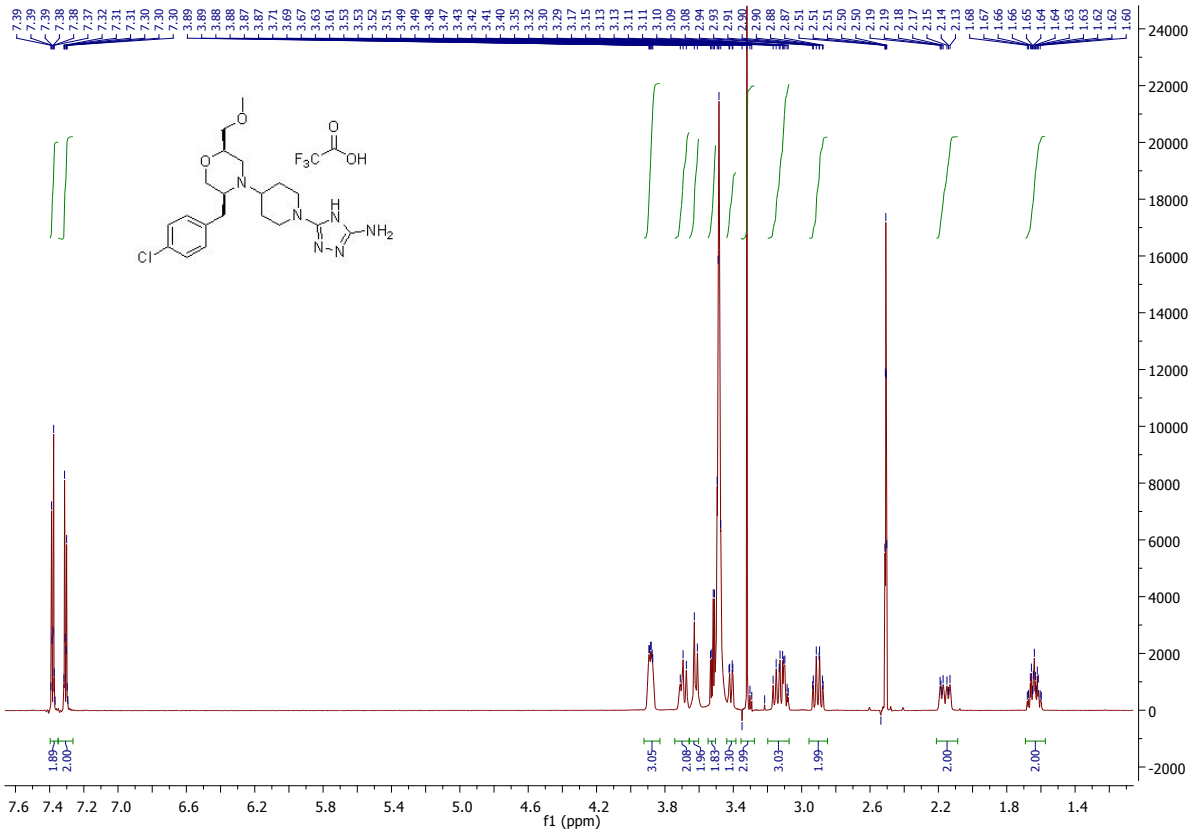
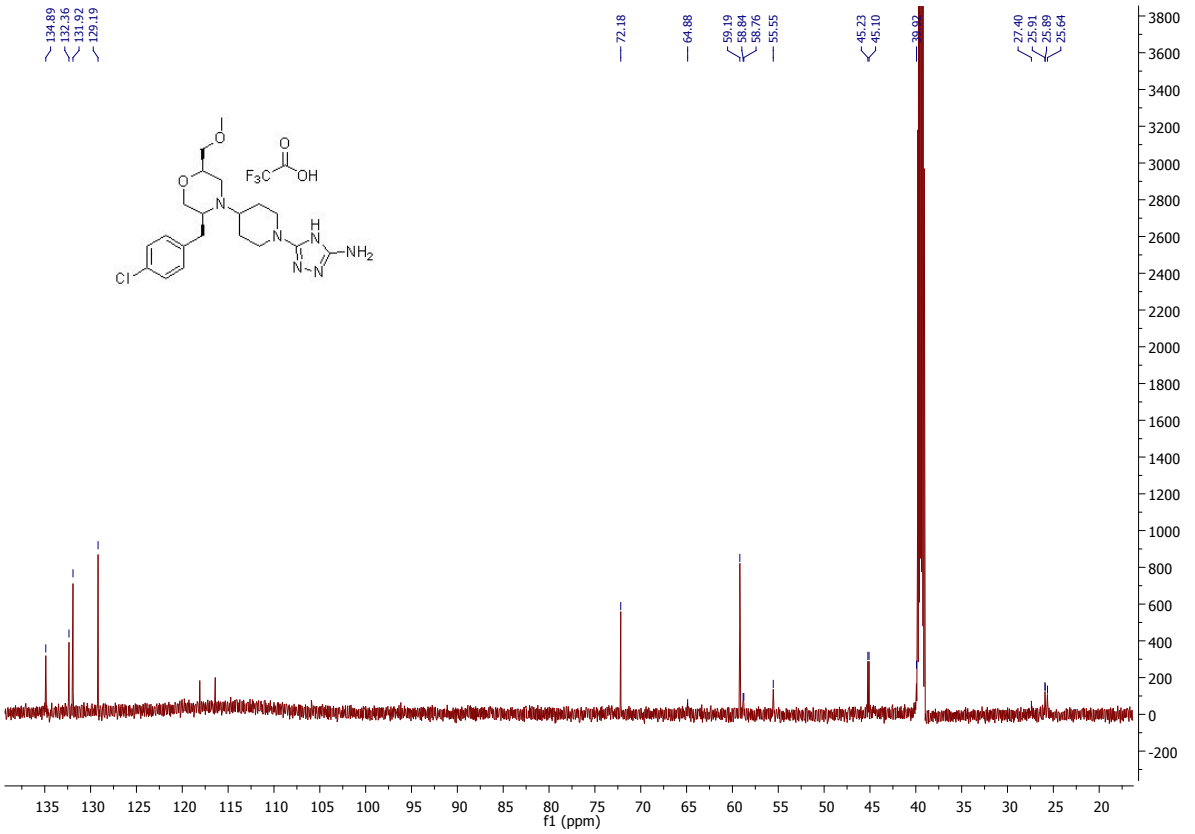
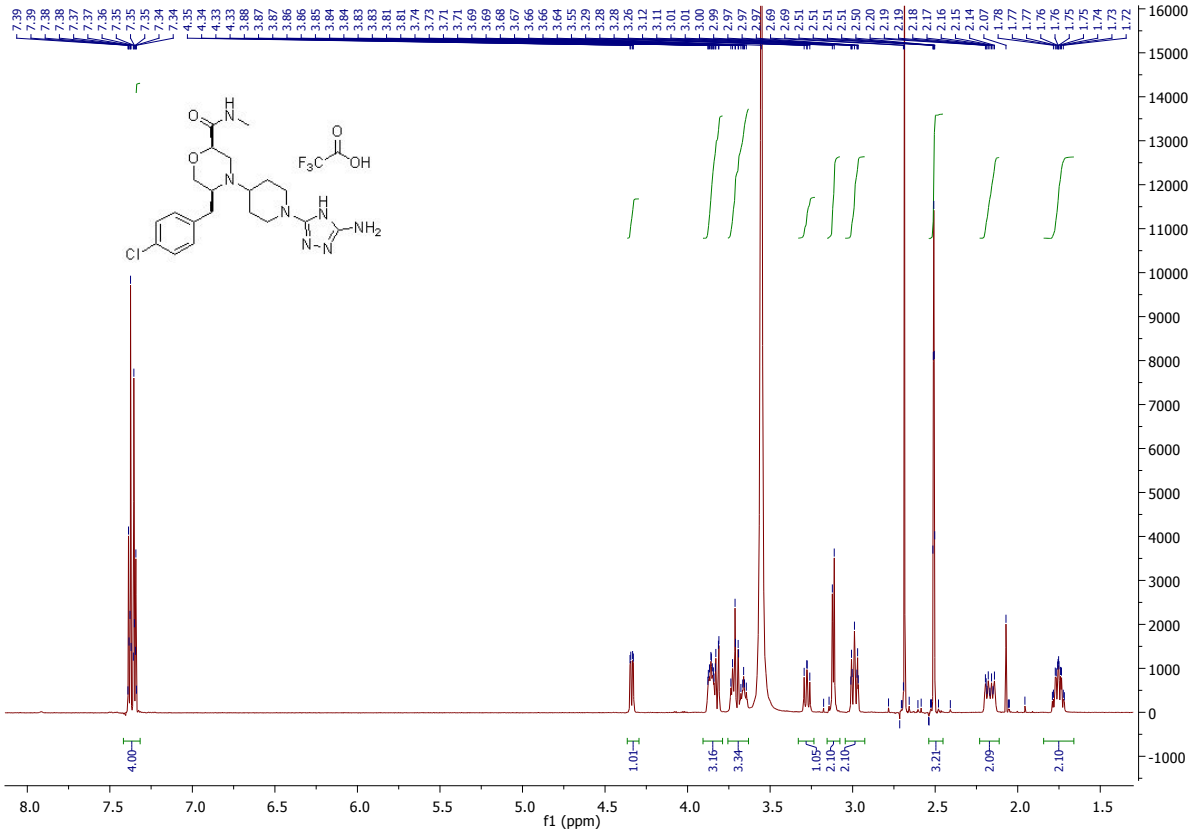
N
H
N
N
NH2
Cl
f1 (ppm)

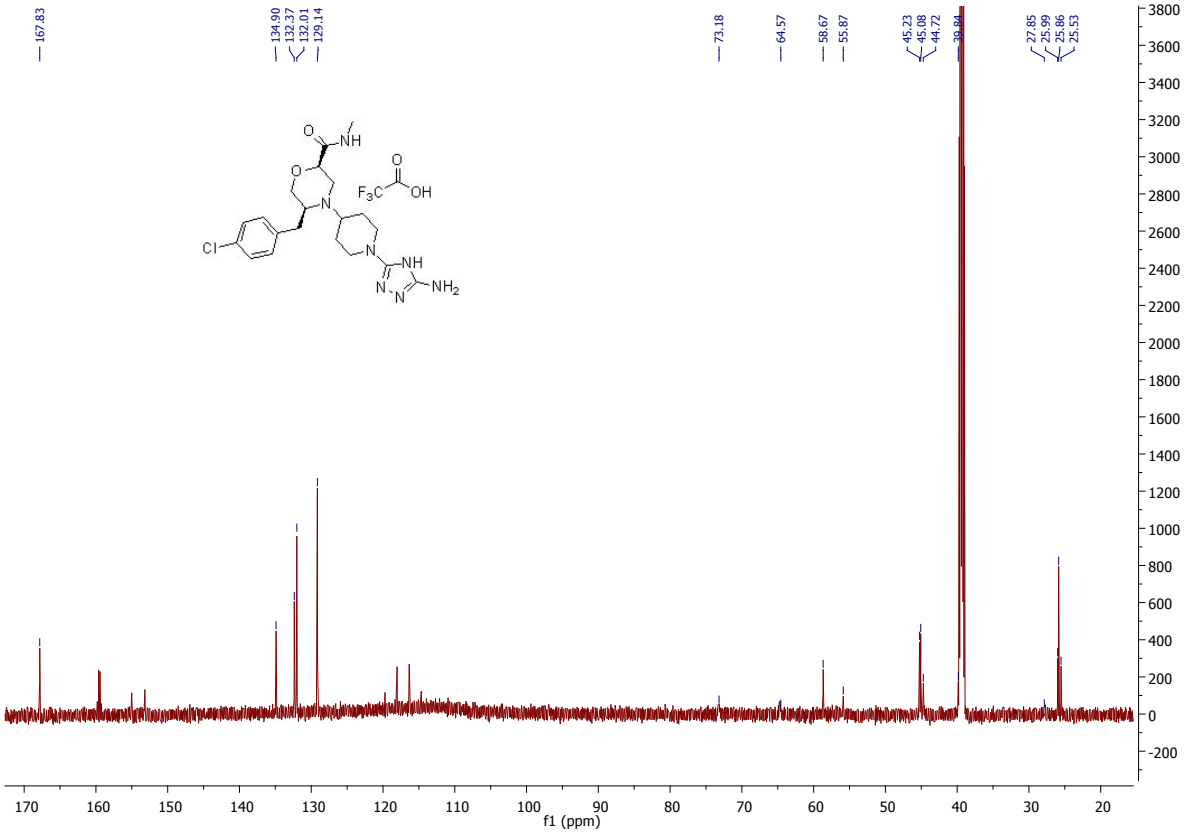
Example 16.



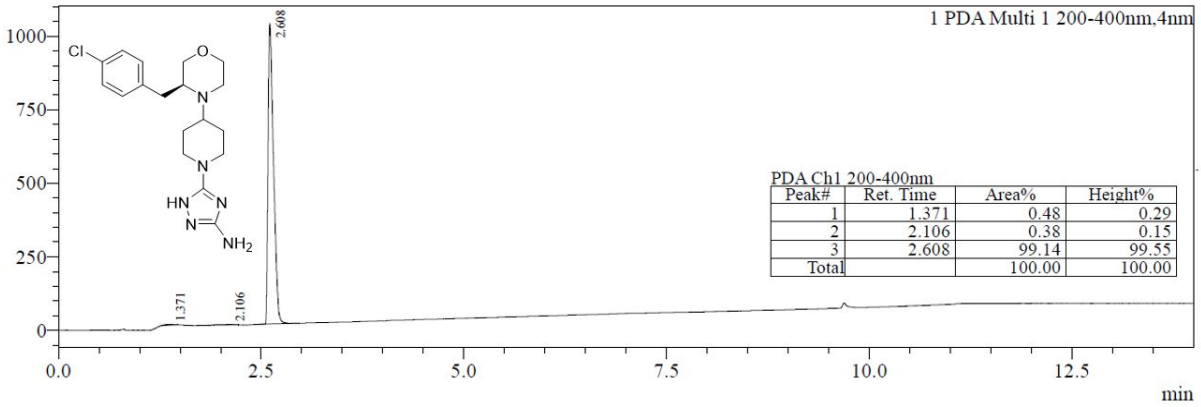


Example 17.





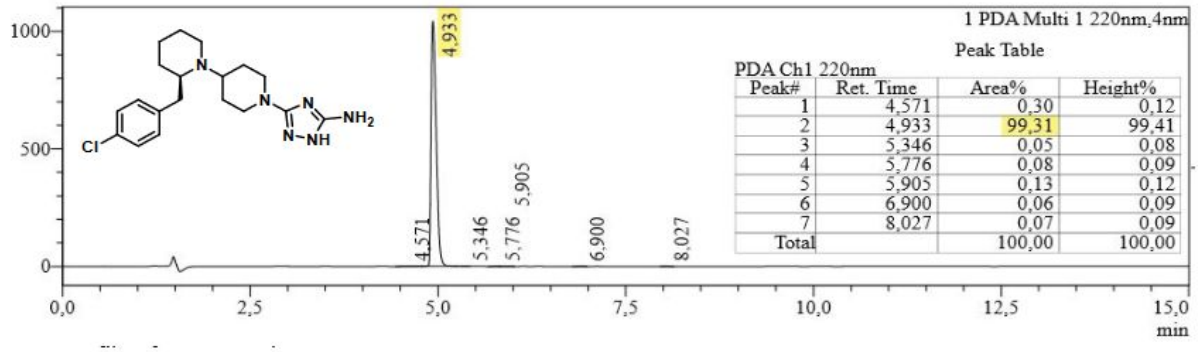
HPLC profile of compound 3.



PDA Ch1 200-400nm

Peak#	Ret. Time	Area%	Height%
1	1.371	0.48	0.29
2	2.106	0.38	0.15
3	2.608	99.14	99.55
Total		100.00	100.00

HPLC profile of compound 4.

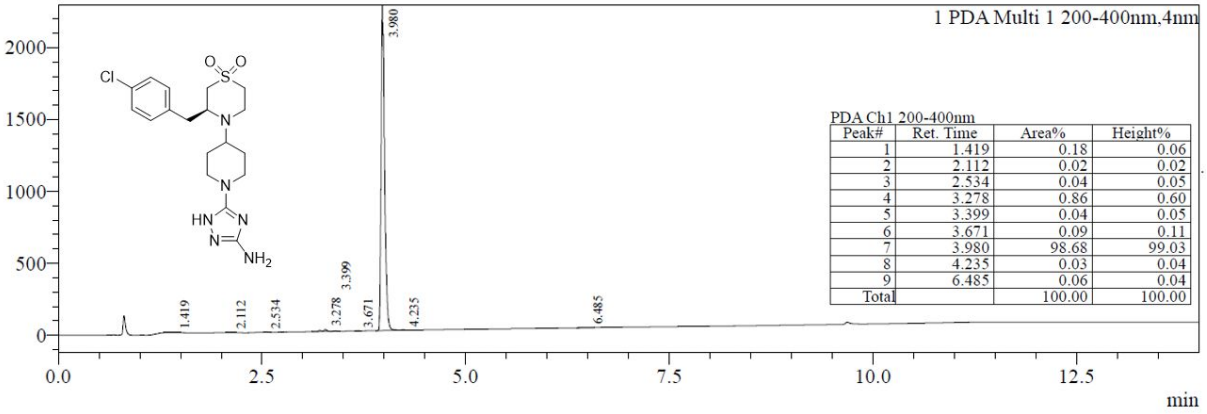


Peak Table

PDA Ch1 220nm

Peak#	Ret. Time	Area%	Height%
1	4,571	0,30	0,12
2	4,933	99,31	99,41
3	5,346	0,05	0,08
4	5,776	0,08	0,09
5	5,905	0,13	0,12
6	6,900	0,06	0,09
7	8,027	0,07	0,09
Total		100,00	100,00

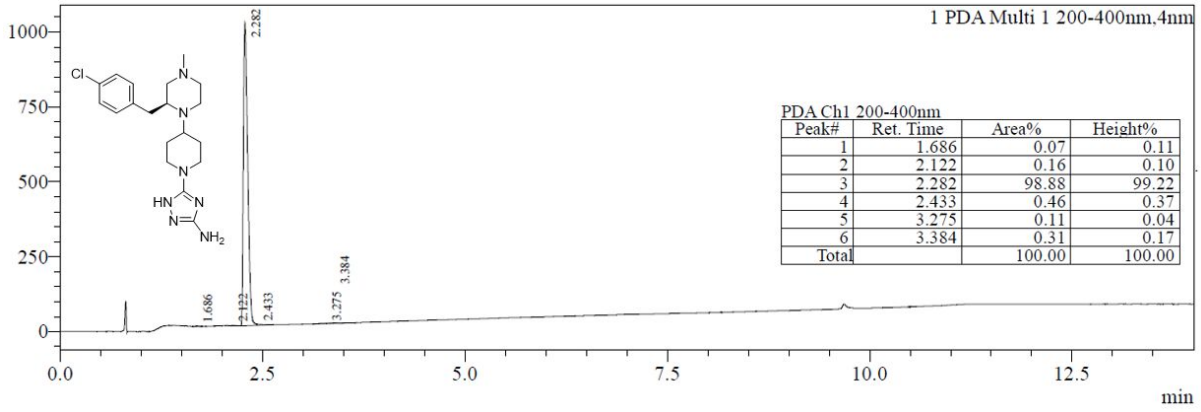
HPLC profile of compound 5.



PDA Ch1 200-400nm

Peak#	Ret. Time	Area%	Height%
1	1.419	0.18	0.06
2	2.112	0.02	0.02
3	2.534	0.04	0.05
4	3.278	0.86	0.60
5	3.399	0.04	0.05
6	3.671	0.09	0.11
7	3.980	98.68	99.03
8	4.235	0.03	0.04
9	6.485	0.06	0.04
Total		100.00	100.00

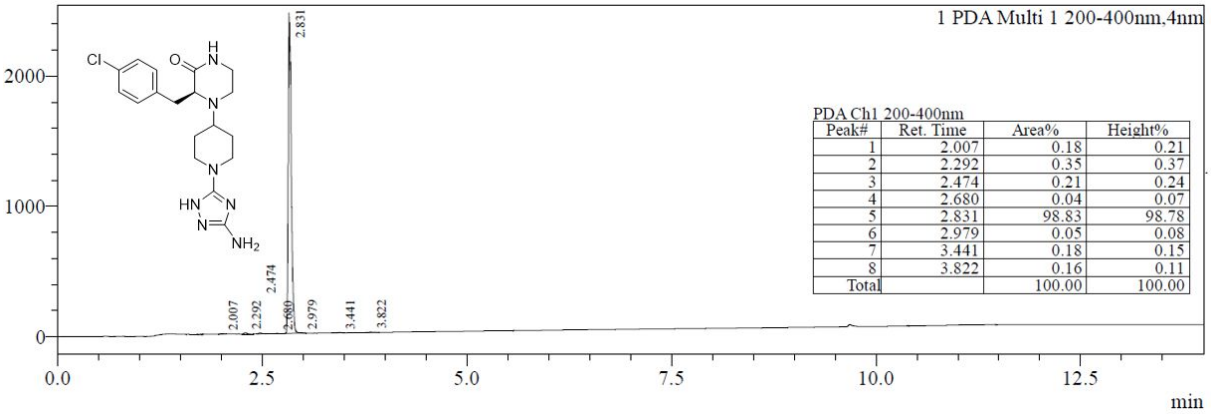
HPLC profile of compound 6.



PDA Ch1 200-400nm

Peak#	Ret. Time	Area%	Height%
1	1.686	0.07	0.11
2	2.122	0.16	0.10
3	2.282	98.88	99.22
4	2.433	0.46	0.37
5	3.275	0.11	0.04
6	3.384	0.31	0.17
Total		100.00	100.00

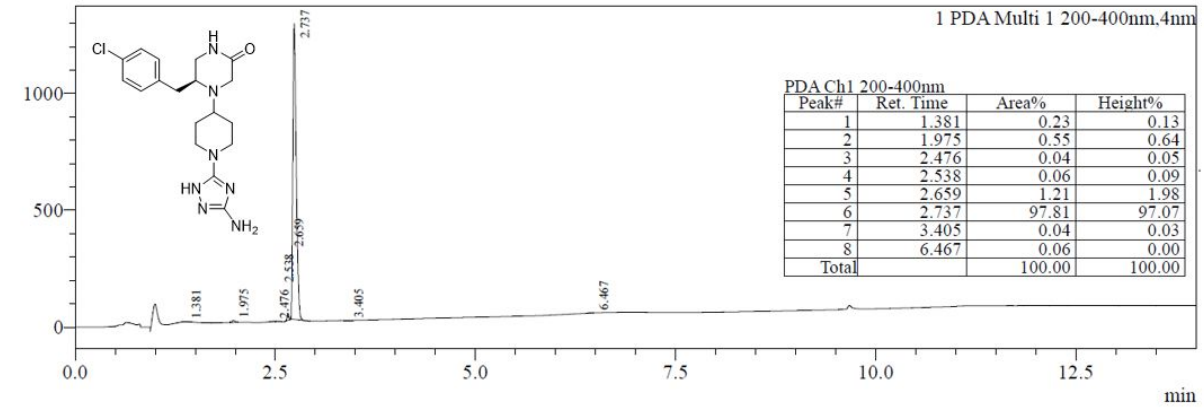
HPLC profile of compound 7.



PDA Ch1 200-400nm

Peak#	Ret. Time	Area%	Height%
1	2.007	0.18	0.21
2	2.292	0.35	0.37
3	2.474	0.21	0.24
4	2.680	0.04	0.07
5	2.831	98.83	98.78
6	2.979	0.05	0.08
7	3.441	0.18	0.15
8	3.822	0.16	0.11
Total		100.00	100.00

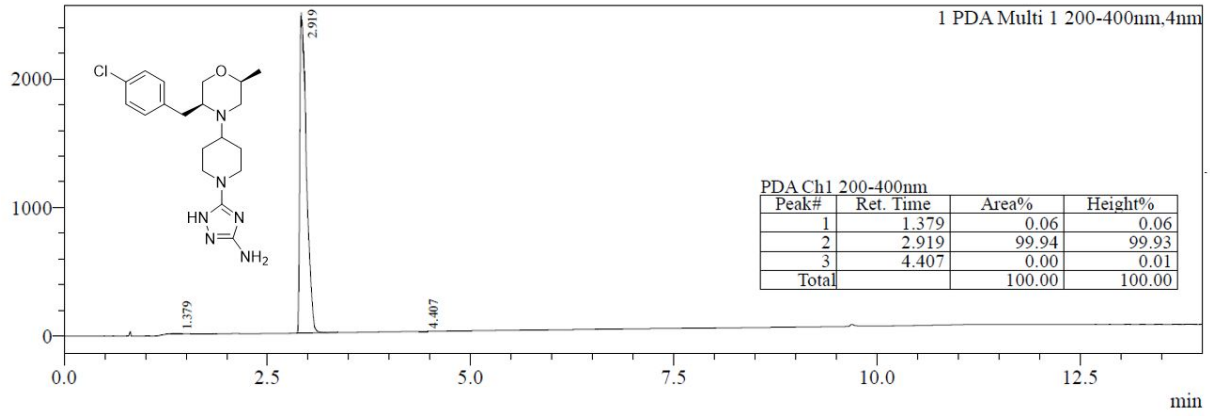
HPLC profile of compound 8.



PDA Ch1 200-400nm

Peak#	Ret. Time	Area%	Height%
1	1.381	0.23	0.13
2	1.975	0.55	0.64
3	2.476	0.04	0.05
4	2.538	0.06	0.09
5	2.659	1.21	1.98
6	2.737	97.81	97.07
7	3.405	0.04	0.03
8	6.467	0.06	0.00
Total		100.00	100.00

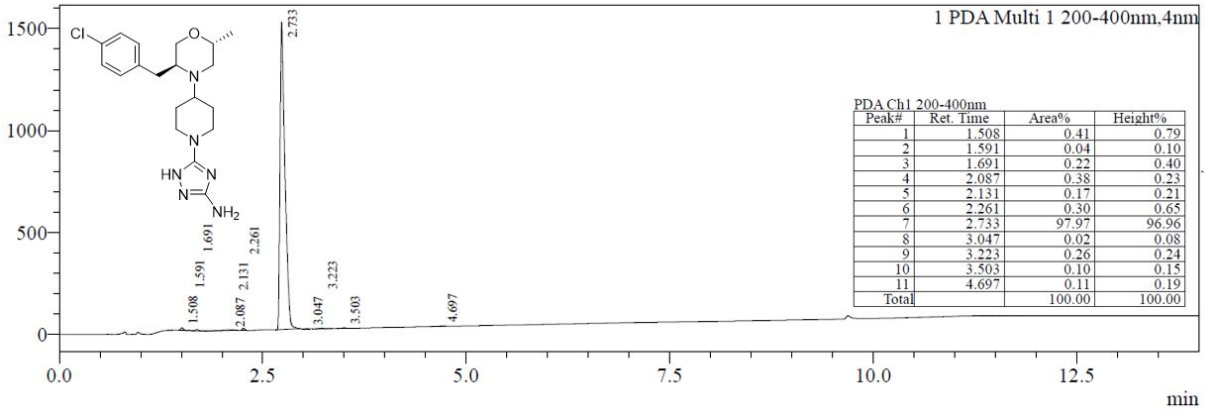
HPLC profile of compound 9.



PDA Ch1 200-400nm

Peak#	Ret. Time	Area%	Height%
1	1.379	0.06	0.06
2	2.919	99.94	99.93
3	4.407	0.00	0.01
Total		100.00	100.00

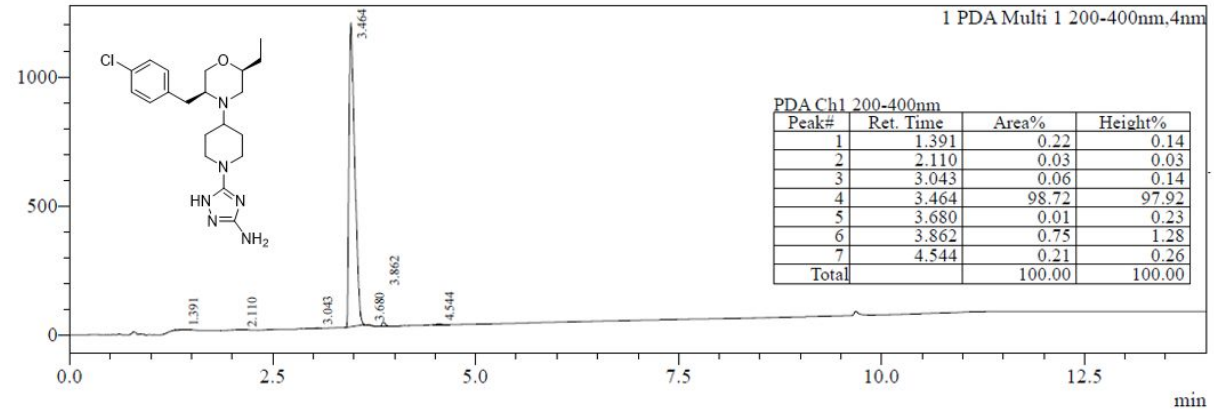
HPLC profile of compound 10.



PDA Ch1 200-400nm

Peak#	Ret. Time	Area%	Height%
1	1.508	0.41	0.79
2	1.591	0.04	0.10
3	1.691	0.22	0.40
4	2.087	0.38	0.23
5	2.131	0.17	0.21
6	2.261	0.30	0.65
7	2.733	97.97	96.96
8	3.047	0.02	0.08
9	3.223	0.26	0.24
10	3.503	0.10	0.15
11	4.697	0.11	0.19
Total		100.00	100.00

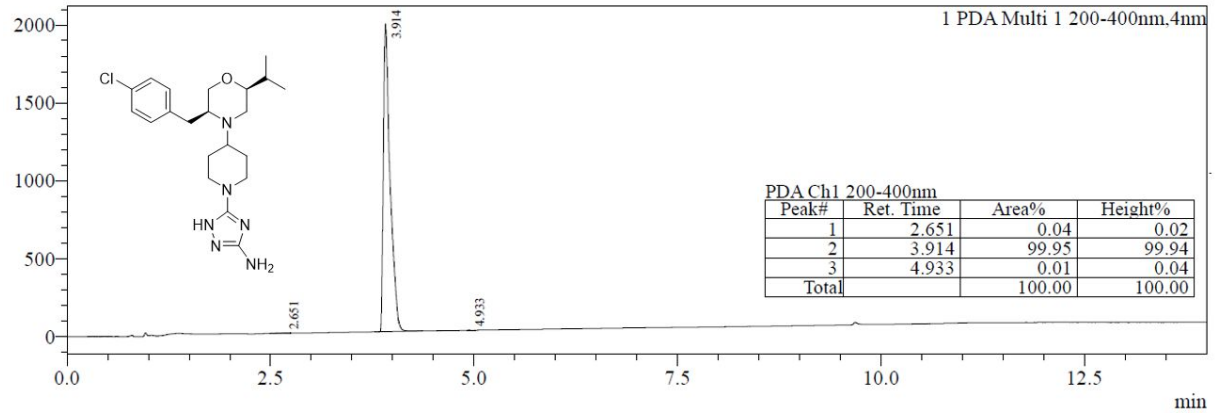
HPLC profile of compound 11.



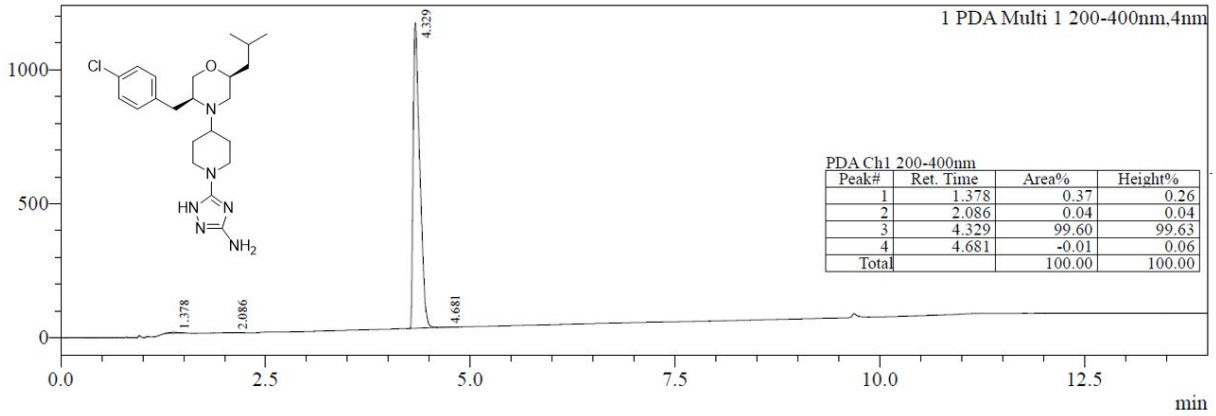
PDA Ch1 200-400nm

Peak#	Ret. Time	Area%	Height%
1	1.391	0.22	0.14
2	2.110	0.03	0.03
3	3.043	0.06	0.14
4	3.464	98.72	97.92
5	3.680	0.01	0.23
6	3.862	0.75	1.28
7	4.544	0.21	0.26
Total		100.00	100.00

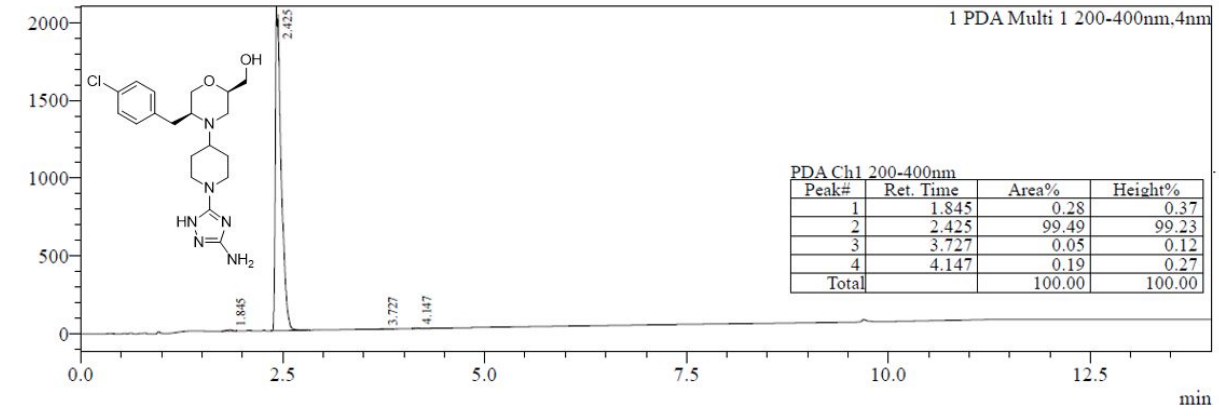
HPLC profile of compound 12.



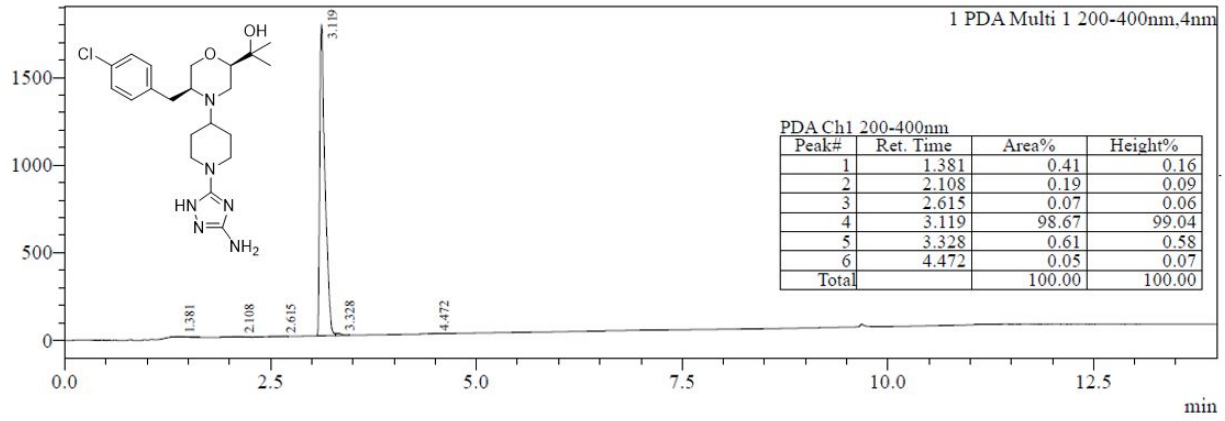
HPLC profile of compound 13.



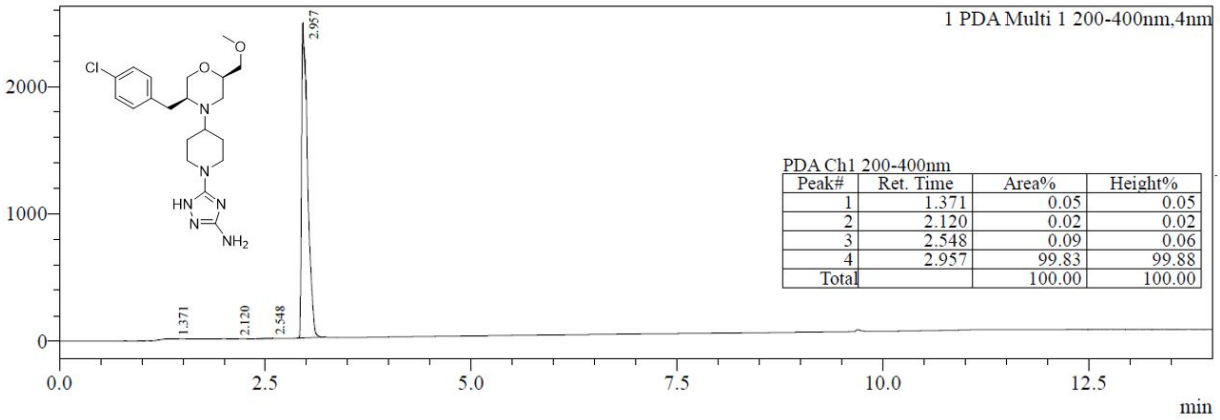
HPLC profile of compound 14.



HPLC profile of compound 15.



HPLC profile of compound 16.



HPLC profile of compound 17.