Discovery of *N*-Alkyl Piperazine Side Chain Based CXCR4 Antagonists with Improved Drug-like Properties

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General Procedures: Unless otherwise indicated, all reactions were conducted in oven (150°C) or flame-dried glassware using distilled and degassed solvents under positive pressure of dry argon with standard Schlenk techniques. Stainless steel syringes or cannulas that had been oven-dried (150°C) and cooled under an argon atmosphere or in a desiccator were used to transfer air- and moisture-sensitive liquids. Yields refer to chromatographically (LC-MS (ESI-API, 254 nm) MeOH in H_2O (0.1% HCO_2H), C18 (Agilent Zorbax XDB-18, 50 mm x 4.6 mm, 3.5 μm), *m/z* and spectroscopically (^{1}H NMR) homogeneous materials, unless otherwise stated. Reactions were monitored by thin-layer chromatography (TLC) carried out on pre-coated glass plates of silica gel (0.25 mm) 60 F_{254} using the indicated solvent system. Visualization was accomplished with ultraviolet light (UV 254 nm), or by shaking the plate in a sealed jar containing silica gel and iodine. Alternatively, plates were treated with one of the following solutions (this was accomplished by holding the edge of the TLC plate with forceps or tweezers and immersing the plate into a wide-mouth jar containing the desired staining solution) and carefully heating with a hot-air gun (450°C) for approximately 1-2 min (NOTE: excess stain was removed by resting the TLC on a paper towel prior to heating): 10% phosphomolybdic acid in ethanol, 1% potassium permanganate/7% potassium carbonate/0.5% sodium hydroxide aqueous solution, and/or anisaldehyde in ethanol with 10% sulfuric acid. Flash column chromatography was performed using Silica Flash® P60 silica gel (40-63 μm) from Silicycle, or Teledyne Isco Combiflash. All work-up and purification procedures were carried out with reagent grade solvents in air.

CXCR4 and Muscarinic Receptors Calcium Flux Assays:

Exemplary compounds were tested for their ability to induce or inhibit calcium flux in CCRF-CEM cells. The experimental procedure and results are provided below. The exemplified biological assays, which follow, have been carried out with all compounds. Human T lymphoblast cells (CCRF-CEM) expressing endogenous CXCR4 receptors and musarinic acetycholine receptors were grown in suspension culture and plated in clear bottom 384-well microplates (Greiner bio-one Cat# 789146) in assay buffer [Hank's Buffered Saline Solution (Gibco Cat# 14025-092) supplemented with 20 mM HEPES (Gibco Cat# 15630-080) and 0.1% fatty-acid free BSA (Sigma Cat# A9205)] at 40,000 cells per well. The cells were loaded with equal volume of calcium indicator dye (AAT Bioquest Inc, Cat# 34601) for 30 minutes at 37°C. The cells were then equilibrated to room temperature for 15 minutes

before assay. Test compounds solubilized and serially diluted in DMSO were transferred to 384 well plates (Matrix Cat# 4307). The serially diluted compounds were diluted to working concentrations with the same assay buffer to 0.5% DMSO. They were added to the cells by FDSS6000 (Hamamatsu) at final concentrations ranging from 25,000 nM to 0.423 nM. Activity of the compounds to induce calcium flux was monitored by FDSS in the "agonist mode" for 90 sec. For "antagonist mode" assessment, the cells are subsequently incubated for 25 minutes at room temperature. SDF-1α (R&D System Cat# 350-NS/CF) or acetylcholine was then added at a final concentration of 5 nM and 2,000 nM, respectively, to stimulate the cells. Inhibition of SDF-1α and acetylcholine-induced calcium flux was monitored by FDSS6000 for 90 seconds.

Activation data for the test compound over a range of concentrations was plotted as percentage activation of the test compound (100% = maximum response triggered by a saturating concentration of SDF-1α, i.e., 160 nM). After correcting for background, EC_{50} values were determined. The EC_{50} is defined as the concentration of test compound, which produces 50% of the maximal response and was quantified using the 4-parameter logistic equation to fit the data. Inhibition data for the test compound over a range of concentrations was plotted as percentage inhibition of the test compound as compared to an internal control compound. The IC_{50} is defined as the concentration of test compound, which inhibits 50% of the maximal response and was quantified using the 4-parameter logistic equation to fit the data.

None of the compounds tested demonstrated agonist activity in the calcium flux assay. All compounds demonstrated EC_{50} values >30 μM. In contrast, compounds demonstrated a range of potencies in inhibiting SDF-1 α -induced calcium flux.

PAMPA Assay:

Compounds and controls are utilized as 10 mM stocks in 100% DMSO. Compounds are diluted 1:100 in pH 7.4 or pH 5.5 donor well buffer (pION CAT # 110151) providing a 100 µM assay solution in 1% DMSO. Compound diluted in donor well buffer is transferred to a Whatman Unifilter plate and filtered prior to dispensing 200 µl into the donor well of the assay plate (pION CAT #110163). The PAMPA membrane is formed by pipetting 4 µl of the lipid solution (pION CAT #110169) onto the filter plate (VWR CAT #13503). The membrane is then covered with 200 µl of acceptor well buffer at pH 7.4 (pION CAT #110139). The PAMPA assay plate (donor side and acceptor side) is combined and allowed to incubate at room temperature for 4 hours. The plate is then disassembled and spectrophotometer plates (VWR CAT #655801) are filled (150 µl/well). The donor, acceptor, reference, and blank plates are read in the SpectraMax UV plate reader. Data is captured by the pION software which analyzes the spectra and generates Pc values.

Recombinant CYP2D6 Inhibition Assay:

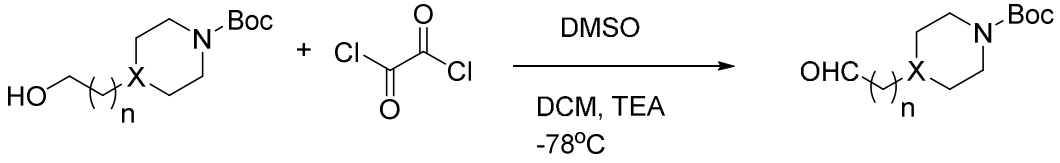
The CYP2D6 inhibition assay utilizes microsomes from the insect cells expressing human recombinant CYP2D6 enzyme and fluorogenic probe (AMMC, 3-[2-(N,N-diethyl-N-methylamino)ethyl]-7-methoxy-4-methylcoumarin) that produces fluorescent metabolite; both reagents were obtrained from Thermo Fisher Scientific/Discovery Labware (Woburn, MA). Assay was performed in 1536-well microplates in a total volume of 5 µl. Automated liquid handling equipment (Thermo Multidrop Combi, LabCyte ECHO 550) was used in all steps of compound preparation and for assay reagent additions. Each compound was tested in duplicate at 7 concentrations ranging from 1 nM to 20 µM; final concentration of DMSO in reactions was 0.2%. Positive controls were included in each experiment/run. Test compounds (10 nL/well) were first pre-incubated at 37°C for 30 min with 2.5 µL of prewarmed 2-fold-concentrated mixture of AMMC fluorogenic substrate (3 µM) and 12.5 nM rCYP2D6 enzyme in 100 mM potassium phosphate assay buffer pH 7.4. At the end of preincubation, the reactions were initiated by the addition of 2.5 µL of prewarmed 2-fold-concentrated NADPH-regenerating system (16.2 nM NADP) in the same assay buffer. Assay plates were then incubated at 37°C for 45 min. Following incubation, reactions were terminated by the addition of 3 µL of quench buffer (80% acetonitrile, 20% 0.5 M TRIS-base). Fluorescence intensity was measured using the Envision fluorescence plate reader (Perkin Elmer) at excitation and emission wavelengths of 405 and 460 nm,

respectively, using a 430-nm cut-off filter. The end-point fluorescence readout was normalized to the fluorescence intensity of the reaction performed in the absence of the test substance (totals, 0% inhibition) and the mixture of reaction components in the presence of "Inhibitor Cocktail" (background, 100% inhibition). The IC_{50} value for each compound is derived from the fitted 20-point curve using a four-parameter logistic regression model.

For Metabolic Stability experimental see Ref. 3.

Experimental:

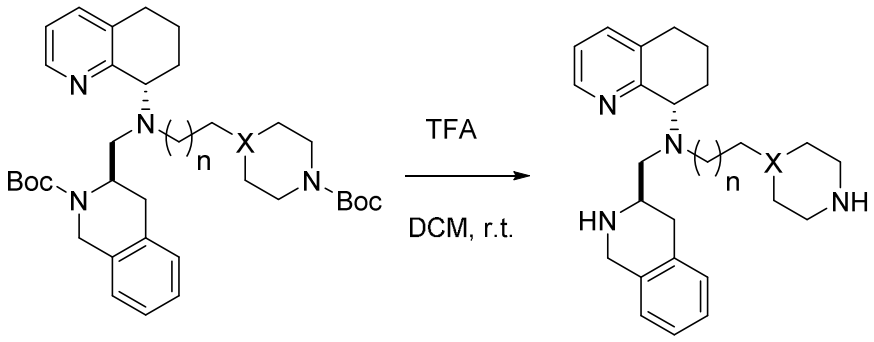
General procedure for corresponding aldehydes (8-10, 18a and **18b):**



A solution of oxalyl chloride (1.23 ml, 14.1 mmol) in DCM (30.9 ml) was cooled to -78° C (dry-ice acetone) under argon atmosphere. DMSO (1.77 ml, 25.1 mmol) was then added dropwise over a period of 35-45 minutes. After the addition was complete, the reaction was stirred for 30 minutes. Corresponding alcohol (6.27 mmol) was then added in one portion while maintaining the temperature below -70° C and the reaction mixture was allowed to stir for 1 hour. Triethylamine (TEA) (3.50 ml, 25.06 mmol) was then added dropwise over a period of 30 minutes. After the addition was complete the mixture was stirred at -78° C for 1 hour and then warmed to 0 °C and stirred for 2 hours. The reaction was quenched by addition of 10 mL saturated $NaHCO_3$ solution. The organic layer was separated and aqueous layer extracted with DCM (2 x 40 mL). The combined organic layers were washed with brine, dried over magnesium sulfate and solvent was removed under reduced pressure to yield light yellow oil (60-95% yield).

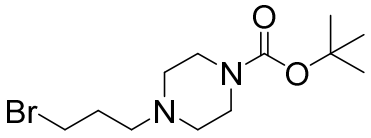
8-10, 18a and **18b** were used without purification

General procedure for Boc deprotection:



Boc protected compound (0.5 mmol) and 1 ml of TFA dissolved in 10 ml DCM were stirred at room temperature for 1-24 hour. The reaction mixture was basified by addition of 1N NaOH to pH>10-12. The aqueous phase was extracted three times with DCM and the combined organic layer was dried over anhydrous $MgSO_4$, filtered, and evaporated. The crude product was purified with silica gel column chromatography using DCM: MeOH: NH_3 gradient.

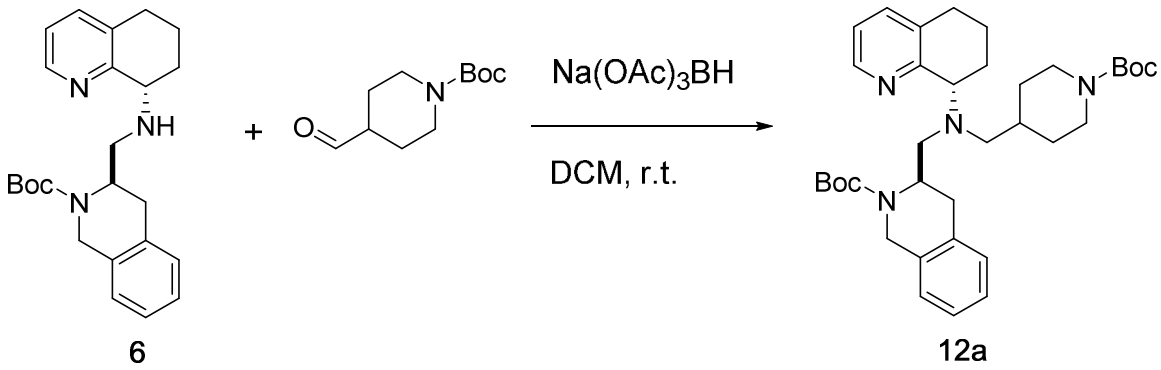
***tert*-butyl-4-(3-bromoprpyl)piperazine-1-carboxylate (11):**



tert-Butyl piperazine-1-carboxylate (5.00 g, 26.8 mmol) was dissolved in dioxane (100 ml) and K_2CO_3 (18.6 g, 134 mmol) and 1,3-dibromopropane (27.2 ml, 268 mmol) were added and the suspension was heated at 65°C for overnight. The reaction mixture was cooled to room temperature and the inorganic by-products were filtered off. The filtrate was evaporated and the residue was purified by silica gel column chromatography using hexanes/ethyl acetate gradient. (4.12 g, 50% yield, yellow oil)

^{1}H NMR (400Hz, $CDCl_3$) δ 1.46 (s, 9H), 2.07-2.02 (m, 2H), 2.39 (brs, 4H), 2.49 (t, *J*= 6.4 Hz, 2H), 3.43 (brs, 4H), 3.47 (t, *J*= 6.4 Hz, 2H).

General procedure for compounds 12a-c, and **12e:**

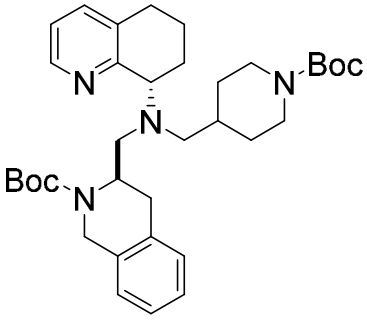


6 12a

(*R*)-*tert*-Butyl-3-(((S)-5,6,7,8-tetrahydroquinolin-8-ylamino)methyl)-3,4-dihydroisoquinoline-2(1H)-carboxylate (**6**) (0.500 g, 1.27 mmol) and *tert*-butyl 4-formylpiperidine-1-carboxylate (0.298 g, 1.40 mmol) were dissolved in DCM (15 mL). Sodium triacetoxyborohydride (STAB (0.404 g, 1.91 mmol) was then added in one portion, followed by addition of acetic acid (0.073 ml, 1.27 mmol). The reaction was allowed to stir under argon overnight. The reaction was quenched by addition of aqueous $NaHCO_3$, extracted with DCM (2 x 100 mL), dried over $MgSO_4$ and evaporated under reduced pressure. The resulting yellow oil was purified by silica gel column chromatography using DCM: MeOH: NH_4OH gradient.

(*R*)-*tert*-butyl-3-((((1-(tert-butoxycarbonyl)piperidin-4-yl)methyl)((*S*)-5,6,7,8-tetrahydroquinolin-8-yl)amino)methyl)-3,4-dihydroisoquinoline-2(1H)-carboxylate (12a):

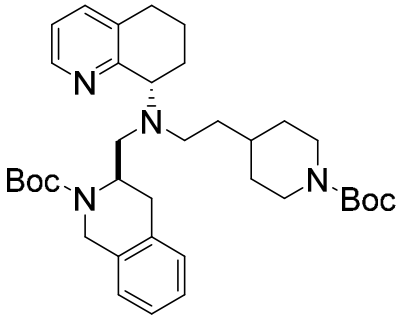
(0.563 g, 75% yield, light yellow oil)



^{1}H NMR (600 MHz, $CDCl_3$) δ 8.46 (d, *J* = 4.2 Hz, 1H), 7.31 (d, *J* = 4.2 Hz, 1H), 7.06-7.02 (m, 5H), 4.18-4.12 (m, 2H), 4.10-3.95 (m, 2H), 3.52-3.12 (m, 4H), 2.96 (d, *J* =11.4 Hz, 2H), 2.80- 2.75 (m, 2H), 2.61-2.56 (m, 5H), 1.98-1.85 (m, 2H), 1.81-157 (m, 6H), 1.50 (s, 9H), 1.45 (s, 9H); LC-MS [(ESI-API, 254 nm) 50-95% MeOH in H_2O

(0.1% HCO_2H), 3 min, 1.00 mL/min, C18 (Agilent Zorbax XDB-18, 50 mm x 4.6 mm, 3.5 µm)]: *m/z* = 591.2 (M+H), t = 1.301 min.; HPLC/MS purity (> 95%) rt = 1.301 at 254nm, 50-95% MeOH over 3 minutes.

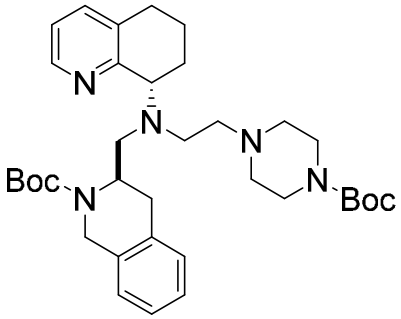
(R)-*tert*-butyl-3-(((2-(1-(*tert*-butoxycarbonyl)piperidin-4-yl)ethyl)-((S)-5,6,7,8-tetrahydroquinolin-8-yl)amino)methyl)-3,4-dihydroisoquinoline-2(1H)-carboxylate (12b):



Used without purification for the next step.

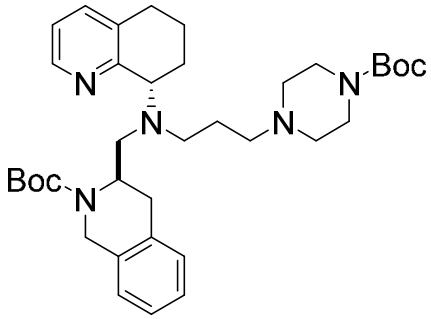
(*R*)-*tert*-Butyl3-(((2-(4-(tert-butoxycarbonyl)piperazin-1-yl)ethyl)((S)-5,6,7,8-tetrahydroquinolin-8-yl)amino)methyl)-3,4-dihydroisoquinoline-2(1H)-carboxylate (12c):

(0.692 g, 90% yield, colorless oil)



^{1}H NMR (400 MHz, CDCl3) δ 8.37 (d, *J* = 12.3 Hz, 1H), 7.26 (s, 1H), 7.14 - 7.08 (m, 1H), 7.03 (d, *J* = 25.1 Hz, 4H), 4.66 (m, 1H), 4.15 (d, *J* = 17.1 Hz, 1H), 3.89 (s, 1H), 3.49 - 3.34 (m, 6H), 3.11 - 2.88 (m, 2H), 2.64 - 2.53 (m, 3H), 2.45 (m, 1H), 2.34 (m, 4H), 2.06 - 1.87 (m, 2H), 1.85 - 1.58 (m, 1H), 1.50 (s, 9H), 1.46 (s, 9H); LC-MS [(ESI-API, 254 nm) 95% MeOH in H_2O (0.1% HCO_2H), 3 min, 1.00 mL/min, C18 (Agilent Zorbax XDB-18, 50 mm x 4.6 mm, 3.5 µm)]: *m/z* = 606.3 (M + H), t = 0.685 min.

(R)-*tert*-Butyl-3-(((3-(4-(*tert*-butoxycarbonyl)piperazin-1-yl)propyl)((S)-5,6,7,8-tetrahydroquinolin-8-yl)amino)methyl)-3,4-dihydroisoquinoline- 2(1H)-carboxylate (12d):



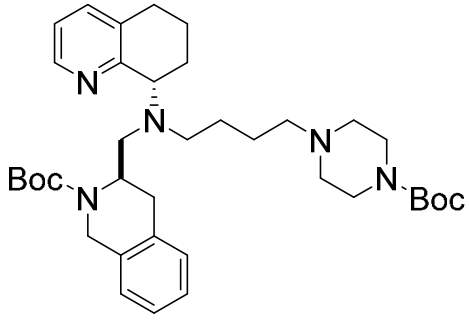
The secondary amine **6** (0.436 g, 1.11 mmol), diisopropylethylamine (DIPEA) (0.387 ml, 2.22 mmol), KI (0.018 g, 0.111 mmol), and **11** (0.511 g, 1.66 mmol) were dissolved in 25 ml of acetonitrile and heated at 65°C for overnight. The reaction mixture was cooled to room temperature and saturated $NaHCO_3$ solution is added. The aqueous phase was extracted with DCM and The combined organic layer was dried over anhydrous $MgSO_4$, filtered and evaporated. The residue was purified by silica gel column chromatography using DCM: MeOH: NH_4OH (90:10:1) as eluent. (0.502 g, 73% yield, white foam)

^{1}H NMR (400 MHz, CDCl3) δ 8.37 (d, *J* = 12.3 Hz, 1H), 7.26 (s, 1H), 7.14 - 7.08 (m, 1H), 7.03 (d, *J* = 25.1 Hz, 4H), 4.66 (m, 1H), 4.15 (d, *J* = 17.1 Hz, 1H), 3.89 (s, 1H), 3.49 - 3.34 (m, 6H), 3.11 - 2.88 (m, 2H), 2.64 - 2.53 (m, 3H), 2.45 (m, 1H), 2.34 (m, 6H), 2.06 - 1.87 (m, 2H), 1.85 - 1.58 (m, 1H), 1.49 (s, 9H), 1.46 (s, 9H).

LC-MS [(ESI-API, 254 nm) 75-95% MeOH in H_2O (0.1% HCO_2H), 3 min, 1.00 mL/min, C18 (Agilent Zorbax XDB-18, 50 mm x 4.6 mm, 3.5 μm)]: *m/z* = 620.6 (M + H), t = 1.485 min.

(R)-*tert*-butyl-3-(((4-(4-(*tert*-butoxycarbonyl)piperazin-1-yl)butyl)((S)-5,6,7,8-tetrahydroquinolin-8-yl)amino)methyl)-3,4-dihydroisoquinoline-2(1H)-carboxylate (12e):

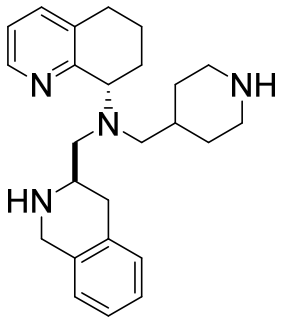
(0.660 g, 82% yield, white foam)



^{1}H NMR (400 MHz, $CDCl_3$) δ 8.37 (d, *J* = 12.3 Hz, 1H), 7.26 (s, 1H), 7.14 - 7.08 (m, 1H), 7.03 (d, *J* = 25.1 Hz, 4H), 4.66 (m, 1H), 4.15 (d, *J* = 17.1 Hz, 1H), 3.89 (s, 1H), 3.49 - 3.34 (m, 6H), 3.11 - 2.88 (m, 2H), 2.64 - 2.53 (m, 3H), 2.45 (m, 1H), 2.34 (m, 6H), 2.06 - 1.87 (m, 2H), 1.85 - 1.58 (m, 3H), 1.49 (s, 9H), 1.46 (s, 9H).

LC-MS [(ESI-API, 254 nm) 95% MeOH in H_2O (0.1% HCO_2H), 3 min, 1.00 mL/min, C18 (Agilent Zorbax XDB-18, 50 mm x 4.6 mm, 3.5 μm)]: *m/z* = 634.5 (M + H), t = 0.897 min.

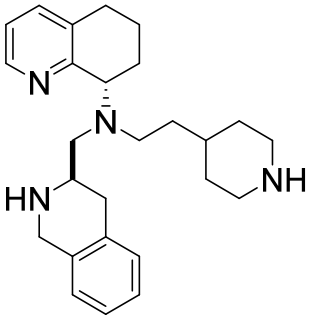
(S)-N-(piperidin-4-ylmethyl)-N-(((R)-1,2,3,4-tetrahydroisoquinolin-3-yl)methyl)-5,6,7,8- tetrahydroquinolin-8-amine (13):



General procedure for Boc deprotection (0.068 g, 35% yield, white foam).

^{1}H NMR (600 MHz, $CDCl_3$) δ 8.42 (d, *J* = 4.2 Hz, 1H), 7.33 (d, *J* = 4.2 Hz, 1H), 7.06-7.02 (m, 5H), 4.08-4.05 (m, 2H), 4.03-3.95 (m, 1H), 3.12 (m, 3H), 2.96 (d, *J* =11.4 Hz, 1H), 2.80- 2.75 (m, 3H), 2.61-2.56 (m, 4H), 2.44-2.41 (m, 2H), 2.32-2.83 (m, 1H), 2.10-2.08 (m, 1H), 1.98-1.85 (m, 4H), 1.79 (d, *J* = 11.4 Hz, 1H), 1.73-1.69 (m, 1H), 1.58-1.57 (m, 1H), 1.27-1.05 (m, 2H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 159.31, 147.09, 136.80, 136.03, 135.14, 134.35, 129.53, 126.91, 126.32, 125.91, 121.77, 62.19, 58.74, 52.92, 49.24, 46.90, 36.69, 34.19, 32.14, 30.67, 29.87, 22.63; HRMS (ESI) $[M+H]^+$, calc'd for $C_{25}H_{35}N_4$ 390.56173, found 390.56284; HPLC/MS purity (> 95%) rt = 1.301 min at 254nm, 50-95% MeOH over 3 minutes.

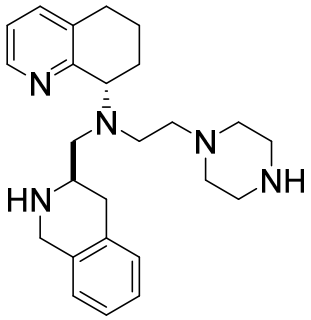
(S)-N-(2-(piperidin-4-yl)ethyl)-N-(((R)-1,2,3,4-tetrahydroisoquinolin-3-yl)methyl)-5,6,7,8- tetrahydroquinolin-8-amine (14):



General procedure for Boc deprotection (0.091 g, 45% yield, white foam).

^{1}H NMR (600 MHz, $CDCl_3$) δ 8.42 (d, *J* = 4.2 Hz, 1H), 7.33 (d, *J* = 4.2 Hz, 1H), 7.06-7.02 (m, 5H), 4.80-4.54 (m, 3H), 4.13-4.04 (m, 2H), 3.95-3.87 (m, 1H), 3.22 (t, *J* =11.4 Hz, 1H), 2.97- 2.88 (m, 4H), 2.83-2.59 (m, 4H), 2.55-2.48 (m, 3H), 2.62-2.04 (m, 1H), 1.98-1.96 (m, 1H), 1.90-1.85 (m, 2H), 1.73-1.67 (m, 2H), 1.63-1.35 (m, 3H), 1.27-1.24 (m, 1H), 1.19-1.15 (m, 1H) ; ^{13}C NMR (100 MHz, $CDCl_3$) δ 159.31, 147.09, 136.80, 136.03, 135.42, 134.58, 129.53, 126.91, 126.32, 125.91, 121.77, 62.19, 58.74, 52.92, 49.24, 46.90, 36.69, 34.19, 31.14, 30.67, 29.87, 22.63; HRMS (ESI) $[M+H]^+$, calc'd for $C_{26}H_{36}N_4$ 404.29754, found 404.29736; HPLC/MS purity (> 95%) rt = 1.324 min at 254nm, 50-95% MeOH over 3 minutes.

(S)-N-(2-(piperazin-1-yl)ethyl)-N-(((R)-1,2,3,4-tetrahydroisoquinolin-3-yl)methyl)- 5,6,7,8-tetrahydroquinolin-8-amine (15):

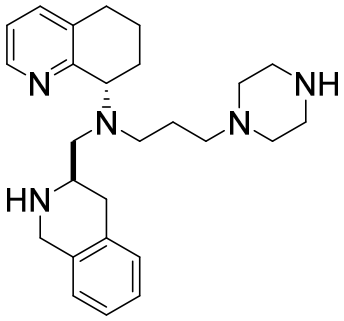


General procedure for Boc deprotection (0.085 g, 42% yield, off white foam).

^{1}H NMR (400 MHz, $CDCl_3$) δ 8.37 (dd, *J* = 6.6, 4.7 Hz, 1H), 7.38 (dd, *J* = 7.3, 5.3 Hz, 1H), 7.19 (m , 2H), 7.15 - 7.03 (m, 3H), 4.53 (dd, *J* = 19.5, 15.6 Hz, 1H), 4.21 (d, *J* = 15.7 Hz, 1H), 4.08 (m, 1H), 3.41 - 3.30 (m, 1H), 3.09 (m, 5H), 2.86 (m, 2H), 2.73 (m, 2H), 2.62 (m, 10H), 2.51 - 2.42 (m, 1H), 2.11 (m, 1H), 2.04 - 1.93 (m, 1H), 1.93 - 1.66 (m, 2H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 157.36, 146.59, 137.64, 137.38, 134.30, 132.05, 131.61, 129.09,

127.69, 126.93, 122.44, 63.87, 56.47, 54.40, 53.77, 52.37, 50.70, 49.95, 43.85, 43.62, 29.62, 29.05; HRMS (ESI) $[M+H]^+$, calc'd for $C_{25}H_{36}N_5$ 406.28925, found 406.28712; HPLC/MS purity (> 95%) rt = 1.320 min at 254nm, 50-95% MeOH over 3 minutes.

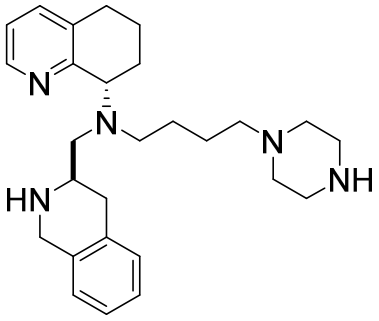
(S)-N-(3-piperazin-1-yl)propyl)-N-(((R)-1,2,3,4-tetrahydroisoquinolin-3-yl)methyl)-5,6,7,8-tetrahydroquinolin-8-amine (16):



General procedure for Boc deprotection (0.100 g, 48% yield, off-white foam).

^{1}H NMR (400 MHz, $CDCl_3$) δ 8.42 (dd, *J* = 4.8, 1.8 Hz, 1H), 7.32 (dd, *J* = 7.7, 1.7 Hz, 1H), 7.11 - 6.96 (m, 5H), 4.07 (dd, *J* = 15.6, 9.1 Hz, 2H), 3.94 (d, *J* = 15.0 Hz, 1H), 3.13 (m, 1H), 2.93 (dd, *J* = 13.3, 3.2 Hz, 1H), 2.88 (t, *J* = 4.9 Hz, 3H), 2.78 (m, 3H), 2.69 - 2.54 (m, 3H), 2.52 - 2.26 (m, 8H), 2.13 - 1.79 (m, 3H), 1.79 - 1.62 (m, 3H), 1.25-1.23 (m, 1H), 1.18-1.16 (m, 1H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 158.76, 146.64, 136.49, 134.59, 133.95, 129.07, 126.41, 125.92, 125.48, 121.36, 117.23, 61.21, 57.57, 57.23, 54.71, 52.34, 48.49, 46.11, 45.18, 29.39, 29.28, 26.73, 21.96; HRMS (ESI) $[M+H]^+$, calc'd for $C_{26}H_{38}N_5$ 420.30490, found 420.30159; HPLC/MS purity (> 95%) rt = 1.340 min at 254nm, 50-95% MeOH over 3 minutes.

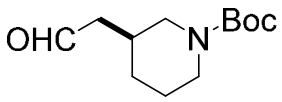
(S)-N-(4-piperazin-1-yl)butyll)-N-(((R)-1,2,3,4-tetrahydroisoquinolin-3-yl)methyl)-5,6,7,8-tetrahydroquinolin-8-amine (17):



General procedure for Boc deprotection (0.091 g, 42% yield, light yellow foam).

^{1}H NMR (400 MHz, $CDCl_3$) δ 8.40 (dd, *J* = 4.7, 1.8 Hz, 1H), 7.37 (dd, *J* = 7.8, 1.8 Hz, 1H), 7.19 - 7.12 (m, 2H), 7.12 - 7.04 (m, 3H), 4.41 - 4.28 (m, 1H), 4.17 - 4.01 (m, 2H), 3.19 - 3.10 (m, 1H), 3.10 - 3.01 (m, 1H), 2.95 (q, *J* = 5.4 Hz, 4H), 2.89 - 2.62 (m, 5H), 2.56-2.46 (m, 5H), 2.30 (dt, *J* = 10.3, 6.6 Hz, 2H), 2.13-2.05 (m, 1H), 2.01-1.93 (m, 1H), 1.88-1.82 (m, 2H), 1.79 - 1.59 (m, 2H), 1.46-1.34 (m, 4H), 1.09 (bs, 1H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 158.20, 146.40, 137.46, 134.21, 132.62, 129.08, 127.07, 126.45, 122.00, 62.90, 58.25, 56.20, 53.00, 51.91, 45.46, 44.98, 44.50, 31.01, 29.19, 27.17, 27.09, 23.91, 21.81; HRMS (ESI) $[M+H]^+$, calc'd for $C_{27}H_{40}N_5$ 434.32055, found 434.32330; HPLC/MS purity (> 95%) rt = 1.350 min at 254nm, 50-95% MeOH over 3 minutes.

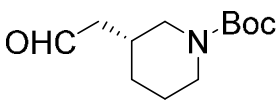
***tert*-Butyl (S)-3-(2-oxoethyl)piperidine-1-carboxylate (18a):**



General procedure for aldehydes (1.60 g, 95%, yellow oil).

^{1}H NMR (400 MHz, $CDCl_3$) δ 9.59 (s, 1H), 3.63-3.61 (m, 2H), 2.71 (bs, 1H), 2.27 (dd, *J* = 3.2 Hz, 2H), 1.88-1.75 (m, 1H), 1.68-1.65 (m, 1H), 1.43-1.41 (m, 1H), 1.25-1.23 (m, 1H), 1.04-1.02 (m, 2H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 200.78, 154.50, 79.15, 49.05, 47.01, 30.41, 28.21, 24.26.

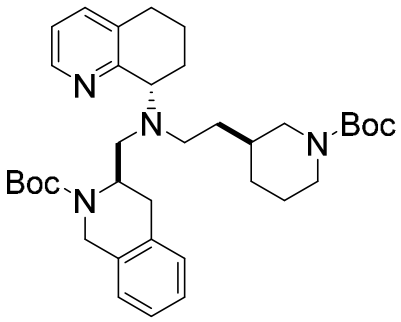
***tert*-Butyl (R)-3-(2-oxoethyl)piperidine-1-carboxylate (18b):**



General procedure for aldehydes (1.60 g, 96%, white goo).

^{1}H NMR (400 MHz, $CDCl_3$) δ 9.58 (s, 1H), 3.64-3.60 (m, 2H), 2.71 (bs, 1H), 2.25 (dd, *J* = 3.2 Hz, 2H), 1.88-1.72 (m, 1H), 1.70-1.65 (m, 1H), 1.43-1.40 (m, 1H), 1.25-1.23 (m, 1H), 1.05-1.02 (m, 2H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 200.78, 154.50, 79.15, 49.05, 47.01, 30.41, 28.21, 24.26.

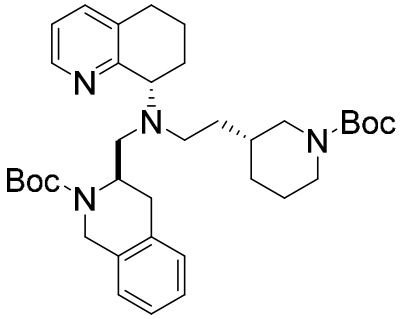
***tert*-butyl (R)-3-(((2-((S)-1-(tert-butoxycarbonyl)piperidin-3-yl)ethyl)((S)-5,6,7,8-tetrahydroquinolin-8-yl)amino)methyl)-3,4-dihydroisoquinoline-2(1H)-carboxylate (19a):**



To the solution of **6** (0.500 g, 1.27 mmol) and **18a** (0.298 g, 1.40 mmol) in 15 ml DCM, STAB (0.404 g, 1.91 mmol) was added in one portion, followed by the addition of acetic acid (0.073 ml, 1.27 mmol). The reaction mixture was allowed to stir under argon overnight. The reaction was quenched by addition of aqueous $NaHCO_3$ solution, extracted with DCM (2 x 100 mL), dried over $MgSO_4$ and evaporated under reduced pressure. The resulting yellow oil was purified by silica gel column chromatography using DCM: MeOH: NH4OH gradient (0.630 g, 82% yield, light yellow foam).

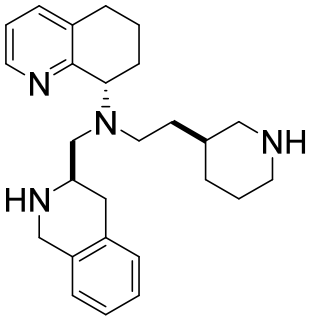
^{1}H NMR (600 MHz, $CDCl_3$) δ 8.34 (d, *J* = 22.5 Hz, 1H), 7.25 – 7.22 (m, 1H), 7.14 – 7.04 (m, 3H), 7.03 – 6.91 (m, 2H), 4.70 – 4.59 (m, 1H), 4.36 (s, 1H), 4.12 (d, *J* = 16.9 Hz, 1H), 3.95 – 3.72 (m, 3H), 3.11 – 2.87 (m, 2H), 2.79 – 2.50 (m, 6H), 2.46 – 2.19 (m, 2H), 1.96 (d, *J* = 45.1 Hz, 2H), 1.78 – 1.51 (m, 4H), 1.48 (s, 9H), 1.41 (s, 9H), 1.37 – 1.23 (m, 3H), 1.16 (d, *J* = 14.8 Hz, 1H), 0.94 (s, 1H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 158.71, 147.06, 136.99, 134.88, 134.35, 134.08, 129.29, 126.69, 126.40, 125.96, 121.87, 62.05, 58.01, 52.74, 51.29, 47.88, 45.92, 34.05, 33.77, 33.35, 30.85, 29.60, 27.69, 24.49, 22.04; HRMS (ESI) $[M+H]^+$, calc'd for $C_{36}H_{53}N_4O_4$ 604.40113, found 604.40650.

***tert*-butyl (R)-3-(((2-((R)-1-(tert-butoxycarbonyl)piperidin-3-yl)ethyl)((S)-5,6,7,8-tetrahydroquinolin-8-yl)amino)methyl)-3,4-dihydroisoquinoline-2(1H)-carboxylate (19b):**



To a solution of **6** (1.30 g, 3.30 mmol) in DCE (35 mL) was added **18b** (0.863 g, 3.30 mmol). To this mixture STAB (1.05 g, 4.96 mmol) was added in one portion. The reaction was kept at ambient temperature and allowed to stir overnight. The reaction mixture was quenched by addition of saturated $NaHCO_3$ solution and the aqueous phase was extracted with DCM (2 x 200 mL). The organic extracts were combined, washed with brine, and dried over MgSO4. The solvent was removed *in vacuo* yielding a light yellow oil. The crude material was used without further purification.

(S)-N-(2-((S)-piperidin-3-yl)ethyl)-N-(((R)-1,2,3,4-tetrahydroisoquinolin-3-yl)methyl)-5,6,7,8-tetrahydroquinolin-8-amine (20):

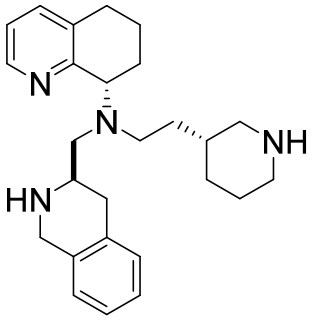


General procedure for Boc deprotection starting with **19a** (0.077 g, 38% yield, off-white foam).

^{1}H NMR (500 MHz, $CDCl_3$) δ 8.45 (d, *J* = 4.8 Hz, 1H), 7.33 (ddt, *J* = 7.7, 1.7, 0.9 Hz, 1H), 7.11 – 7.07 (m, 2H), 7.07 – 7.01 (m, 3H), 4.10 – 4.05 (m, 2H), 3.92 (d, *J* = 15.0 Hz, 1H), 3.15 – 3.08 (m, 1H), 3.07 – 2.99 (m, 2H), 2.94 – 2.75 (m, 5H), 2.71 – 2.62 (m, 3H), 2.59 – 2.52 (m, 1H), 2.43 (dd, *J* = 16.1, 11.0 Hz, 1H), 2.32 (ddd, *J* = 30.6, 12.6, 10.3 Hz, 2H), 2.13 – 2.04 (m, 1H), 2.04 – 1.95 (m, 1H), 1.95 – 1.79 (m, 2H), 1.79 – 1.62 (m, 2H), 1.62 – 1.53 (m, 1H), 1.52 – 1.33 (m, 3H), 1.02 (m, *J* = 12.6, 11.1, 3.9 Hz, 1H); ^{13}C NMR (126 MHz, $CDCl_3$) δ 158.77, 146.72, 136.46, 135.63, 134.66, 133.96, 129.08, 126.46, 125.88, 125.46, 121.37, 60.97, 57.77, 52.96, 52.32, 51.42, 48.73,

46.78, 34.80, 34.37, 33.82, 31.31, 29.44, 29.22, 26.28, 21.98; HRMS (ESI) $[M+H]^+$, calc'd for $C_{26}H_{37}N_4$ 405.30127, found 405.30088; HPLC/MS purity (> 95%) rt = 1.405 at 254nm, 50-95% MeOH over 3 minutes.

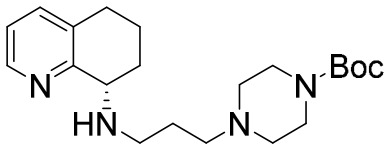
(S)-N-(2-((R)-piperidin-3-yl)ethyl)-N-(((R)-1,2,3,4-tetrahydroisoquinolin-3-yl)methyl)-5,6,7,8-tetrahydroquinolin-8-amine (21):



General procedure for Boc deprotection starting with **19b** (0.081 g, 40% yield, off-white foam).

^{1}H NMR (500 MHz, $CDCl_3$) δ 8.47 (d, *J* = 4.2 Hz, 1H), 7.33 (d, *J* = 7.7 Hz, 1H), 7.10 – 7.07 (m, 2H), 7.07 – 6.99 (m, 3H), 4.10 – 4.03 (m, 2H), 3.86 (d, *J* = 15.1 Hz, 1H), 3.68 – 3.39 (m, 3H), 3.05 (t, *J* = 8.6 Hz, 2H), 3.02 – 2.92 (m, 2H), 2.77 – 2.59 (m, 6H), 2.44 – 2.33 (m, 3H), 2.12 – 2.04 (m, 1H), 2.03 – 1.95 (m, 1H), 1.94 – 1.85 (m, 1H), 1.81 – 1.69 (m, 2H), 1.69 – 1.62 (m, 1H), 1.62 – 1.54 (m, 1H), 1.50 – 1.46 (m, 1H), 1.41 – 1.31 (m, 1H), 1.12 – 1.00 (m, 1H); ^{13}C NMR (126 MHz, $CDCl_3$) δ 158.59, 146.77, 136.54, 135.53, 134.56, 133.87, 129.06, 126.44, 125.89, 125.48, 121.42, 61.42, 57.87, 52.40, 52.03, 51.83, 48.58, 46.37, 34.46, 33.73, 31.26, 29.43, 28.46, 25.55, 21.97. HRMS (ESI) $[M+H]^+$, calc'd for $C_{26}H_{37}N_4$ 405.30127, found 405.30094; HPLC/MS purity (> 95%) rt = 1.419 at 254nm, 50-95% MeOH over 3 minutes.

***tert*-butyl (S)-4-(3-((5,6,7,8-tetrahydroquinolin-8-yl)amino)propyl)piperazine-1-carboxylate (23)**

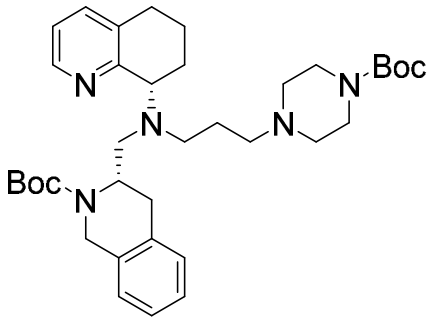


(*S*)-5,6,7,8-tetrahydroquinolin-8-amine (0.941 g, 6.35 mmol) **22**, **11** (1.5 g, 4.88 mmol) and DIPEA (2.13 ml, 12.2 mmol) were dissolved in 20 mL acetonitrile and the reaction mixture was stirred overnight. The reaction mixture was diluted with EtOAc; extracted with saturated aqueous $NaHCO_3$ solution, and water, dried over anhydrous

$MgSO_4$, filtered and evaporated. It was purified by silica gel column chromatography using DCM: MeOH: NH_4OH (9:1:0.1) as eluent (1.78 g, 75% yield, yellow oil).

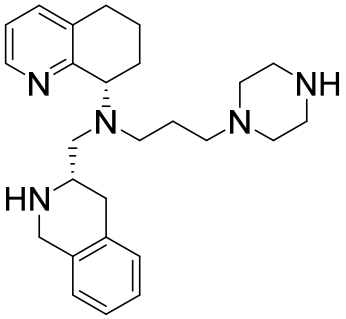
^1H NMR (400 MHz, $CDCl_3$) δ 8.35 (dd, *J* = 4.8, 1.5 Hz, 1H), 7.41 - 7.32 (m, 1H), 7.05 (dd, *J* = 7.7, 4.7 Hz, 1H), 3.77 (d, *J* = 8.2 Hz, 1H), 3.49 - 3.39 (m, 4H), 2.77 (m, 4H), 2.38 (d, *J* = 5.0 Hz, 4H), 2.32 - 2.23 (m, 2H), 2.23 - 2.05 (m, 1H), 2.05 - 1.87 (m, 2H), 1.83 - 1.67 (m, 4H), 1.43 (s, 9H); LC-MS [(ESI-API, 254 nm) 75-95% MeOH in H_2O (0.1% HCO_2H), 3 min, 1.00 mL/min, C18 (Agilent Zorbax XDB-18, 50 mm x 4.6 mm, 3.5 µm)]: *m/z* = 375.3 (M + H), t = 0.820 min.

(S)-*tert*-butyl 3-(((3-(4-(tert-butoxycarbonyl)piperazin-1-yl)propyl)((S)-5,6,7,8-tetrahydroquinolin-8-yl)amino)methyl)-3,4-dihydroisoquinoline-2(1H)-carboxylate (25)



To the solution of *tert*-butyl (*S*)-4-(3-((5,6,7,8-tetrahydroquinolin-8-yl)amino)propyl)piperazine-1-carboxylate (0.300 g, 0.801mmol) **23** in 10 mL DCE, (*S*)-tert-butyl 3-formyl-3,4-dihydroisoquinoline-2(1H)-carboxylate (0.209 g, 0.801 mmol) and STAB (0.340 g, 1.60 mmol) were added and the suspension was stirred at room temperature overnight. The reaction mixture was quenched by addition of saturated aqueous $NaHCO_3$ solution. The aqueous phase was extracted with DCM; the combined organic layers were extracted with water, dried over anhydrous $MgSO_4$, filtered and evaporated. The crude product was used without purification for the next step.

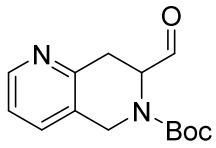
(S)-N-(3-(piperazin-1yl)propyl)-N-(((S)-1,2,3,4-tetrahydroisoquinolin-3-yl)methyl)-5,6,7,8-tetrahydroquinolin-8-amine (26)



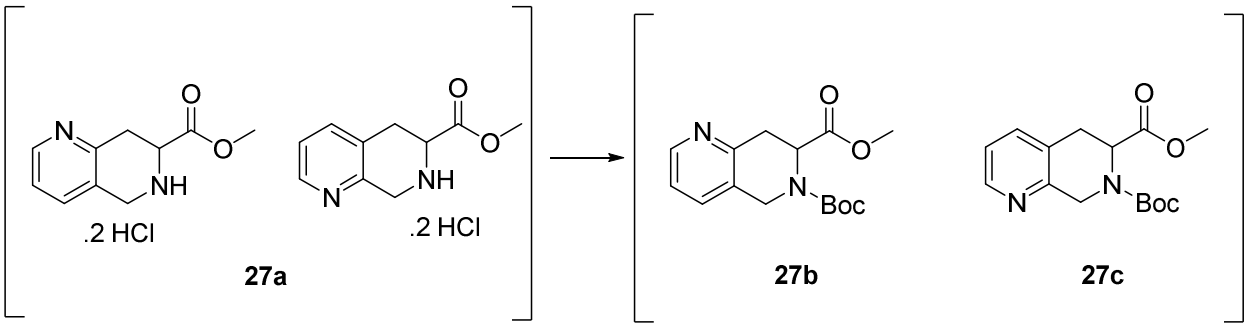
General procedure for Boc deprotection starting with **25** (0.178 g, 85% yield, yellow oil).

^{1}H NMR (500 MHz, $CDCl_3$) δ 8.39 (dd, *J* = 4.8, 1.7 Hz, 1H), 7.35 (dd, *J* = 7.1, 1.6 Hz, 1H), 7.16 - 7.08 (m, 5H), 4.35 (dd, *J* = 19.8, 15.2 Hz, 1H), 4.22 (d, *J* = 15.2 Hz, 1H), 4.01-3.91 (m, 1H), 3.57 (t, *J* = 8.2 Hz, 1H), 2.97 (t, *J* = 5.1 Hz, 3H), 2.93 - 2.89 (m, 1H), 2.89 - 2.79 (m, 2H), 2.78 - 2.70 (m, 3H), 2.65 - 2.47 (m, 5H), 2.46-2.33 (m, 3H), 2.24 - 2.07 (m, 1H), 2.08 - 1.92 (m, 1H), 1.83 - 1.51 (m, 6H), 1.19-1.09 (m, 1H); ^{13}C NMR (126 MHz, $CDCl_3$) δ 157.42, 146.83, 137.53, 134.48, 132.83, 128.75, 127.57, 126.73, 126.67, 122.13, 109.98, 59.82, 56.01, 52.89, 51.93, 51.36, 50.60, 48.58, 44.46, 28.88, 25.52, 23.03, 21.63; HRMS (ESI) $[M+H]^+$, calc'd for $C_{26}H_{38}N_5$ 420.31217, found 420.31259; HPLC/MS purity (> 95%) rt = 1.340 min at 254nm, 50-95% MeOH over 3 minutes.

***tert*-butyl 7-formyl-7,8-dihydro-1,6-naphthyridine-6(5H)-carboxylate (27):**



Step 1:



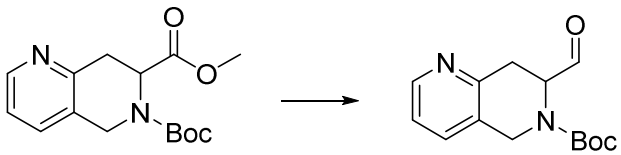
A 1 L rb flask equipped with a magnetic stir bar and rubber septum was charged with 26.9 g of the ester **27a** [1-2] (101 mol, 1 equiv), 42.2 mL of triethylamine (303 mmol, 3 equiv), 1.23 g of DMAP (10.1 mmol, 0.1 equiv) and 253 mL

of THF. Then 27.6 g of Boc_2O (126 mmol, 1.25 equiv) was added and the suspension was stirred for 3 h. The suspension was not going into solution and another portion of 21 mL of TEA (151 mmol, 1.5 equiv) was added followed by 300 mL of acetonitrile and 100 mL of MeOH. After the clear solution was stirred at rt for 12 h, the reaction mixture was concentrated and ethyl acetate was added. The ammonium salts were separated by filtration and the organics were concentrated under vacuum. The crude material was purified on silica gel column using 0-65 % ethyl acetate in hexanes as eluent affording 2.24 g (8 % over 3 steps) of **27c** as a yellow oil and 8.12 g (28 % over 3 steps) of **27b** as a yellow oil which crystallizes in freezer to white solid.

For **27b** (1:1 mixture of conformers)

^{1}H NMR (400 MHz, $CDCl_3$) δ: 8.42 (d, J = 4.9 Hz, 1H), 7.44 (d, J = 7.9 Hz, 0.5H), 7.41 (d, J = 7.7 Hz, 0.5H), 7.14 (dd, J = 7.8, 4.9 Hz, 1H), 5.32 (d, J = 6.9 Hz, 0.5H), 5.01 (dd, J = 6.5, 3.7 Hz, 0.5H), 4.80 (A of AB, J_{AB} = 17.4 Hz, 0.5H), 4.76 (B of AB, J_{AB} = 17.4 Hz, 0.5H), 4.57 (B of AB, J_{AB} = 17.0 Hz, 0.5H), 4.50 (d, J = 16.7 Hz, 1H), 3.65 (s, 1.5H), 3.63 (s, 1.5H), 3.47 (A of AB, J_{AB} = 16.8 Hz, 0.5H), 3.40 (A of ABX, J_{AB} = 17.1 Hz, J_{AX} = 3.9 Hz, 0.5H), 3.32 (B of ABX, J_{AB} = 17.5 Hz, J_{BX} = 6.6 Hz, 1H), 1.53 (s, 4.5H), 1.47 (s, 4.5H). LC-MS [(ESI-API, 254 nm) 95% MeOH in H_2O (0.1% HCO_2H), 3 min, 1.00 mL/min, C18 (Agilent Zorbax XDB-18, 50 mm x 4.6 mm, 3.5 µm)]: *m/z* = 293.1 $(M+H)^+$, 315.0 (M + Na), t = 0.560 min.

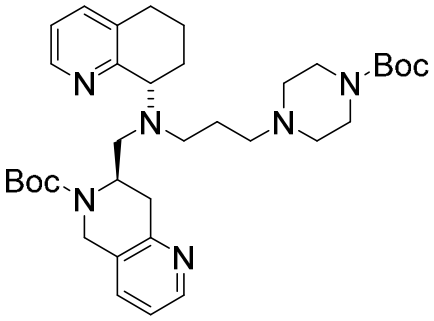
Step 2:



27b **27**

A 100 mL rb flask equipped with a stir bar and rubber septum was charged with 1.00 g of the ester **27b** (3.42 mmol, 1 equiv) and 17.1 mL of toluene. After the reaction mixture was cooled to –78°C, 5.70 mL of 1.2 M solution of DIBAL-H (6.84 mL, 2 equiv) was added dropwise and the solution was stirred at –78°C for 2 h. Then the reaction mixture was quenched by addition of 2.5 mL of MeOH followed by sat. solution of Rochelle's salt. After stirring at rt for 30 min, the product was extracted with EA (3x) and dried over Na_2SO_4. The crude product **27** (994 mg) was used in the next step without purification.

(R)-*tert*-butyl-7-(((3-(4-(*tert*-butoxycarbonyl)piperazin-1-yl)propyl)((S)-5,6,7,8-tetrahydroquinolin-8-yl)amino)methyl)-7,8-dihydro-1,6-naphthyridine-6(5*H*)-carboxylate (28) and **(S)-*tert*-butyl-7-(((3-(4-(*tert*-butoxycarbonyl)piperazin-1-yl)propyl)((S)-5,6,7,8-tetrahydroquinolin-8- yl)amino)methyl)-7,8-dihydro-1,6-naphthyridine-6(5*H*)-carboxylate (30):**



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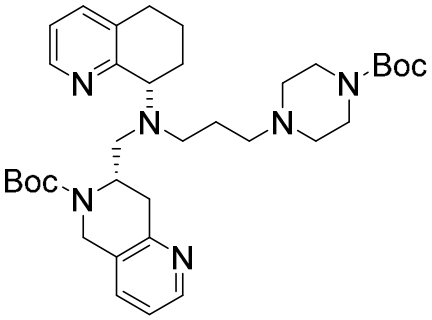


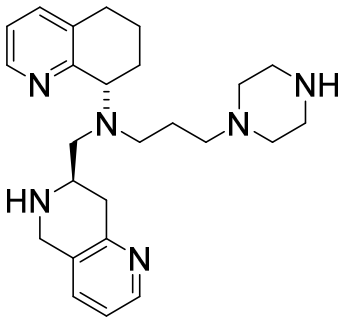
30

A 50 mL Schlenk tube equipped with a stir bar and rubber septum was charged with 262 mg of the *tert*-butyl-7-formyl-7,8-dihydro-1,6-naphthyridine-6(5*H*)-carboxylate (**27**) (0.998 mmol, 1.15 equiv), 325 mg of the amine **23** (0.868 mmol, 1 equiv) and 8.7 mL of CH_2Cl_2. Then 381 µL of titanium isopropoxide (1.30 mmol, 1.5 equiv) was added. After stirring at room temperature for 1 h, 368 mg of STAB (1.74 mmol, 2 equiv) was added and the reaction mixture was stirred at room temperature for 12 h. The reaction mixture was quenched by addition of diluted aqueous $NaHCO_3$ solution, extracted with CH_2Cl_2 (3x) and dried over Na_2SO_4. The crude product is purified on silica gel column using 0 to 10 % MeOH in EtOAc as eluent affording 138 mg (26 %) of **28** and 86 mg (16 %) of **30**.

(*R*)-tert-butyl-7-(((3-(4-(*tert*-butoxycarbonyl)piperazin-1-yl)propyl)((*S*)-5,6,7,8-tetrahydroquinolin-8-yl)amino)methyl)-7,8-dihydro-1,6-naphthyridine-6(5*H*)-carboxylate (28) :

^{1}H NMR (400 MHz, $CDCl_3$) δ: 8.36 (d, *J* = 4.8 Hz, 1H), 8.28 (br s, 1H), 7.27 (d, *J* = 7.4 Hz, 1H), 7.23 (d, *J* = 8.0 Hz, 1H), 7.04 (dd, *J* = 7.7, 4.8 Hz, 1H), 6.93 (br s, 1H), 4.81–4.62 (m, 1.5H), 4.55 (br s, 0.5H), 4.15–3.97 (m, 1H), 3.84 (br s, 1H), 3.59 (br s, 0.5H), 3.36 (s, 4H), 3.46–3.01 (m, 2H), 2.88 (br s, 0.5H), 2.80–2.50 (m, 3H), 2.44–2.35 (m, 1H), 2.28 (s, 7H), 2.07–1.96 (m, 1H), 1.91 (br s, 1H), 1.76–1.37 (m, 4H), 1.46 (s, 9H), 1.42 (s, 9H). LC-MS [(ESI-API, 254 nm) 75-95% MeOH in H2O (0.1% HCO2H), 6 min, 1.00 mL/min, C18 (Agilent Zorbax XDB-18, 50 mm x 4.6 mm, 3.5 µm)]: m/z = 621.2 $(M+H)^+$, t = 0.523 min.

(*S*)-N-(3-(piperazin-1yl)propyl)-N-(((*R*)-5,6,7,8-tetrahydro-1,6-naphthyridin-7-yl)methyl)-5,6,7,8-tetrahydroquinolin-8-amine (29)



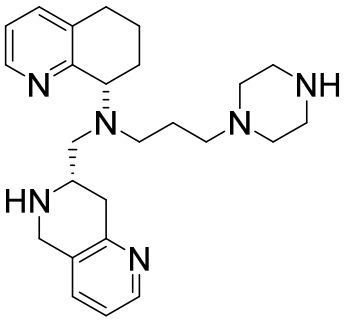
General procedure for Boc deprotection starting with **28** (0.088 g, 94% yield, light yellow foam).

^{1}H NMR (400 MHz, $CDCl_3$, ppm) δ: 8.39 (dd, J = 4.8, 1.7 Hz, 1H), 8.34 (dd, J = 4.8, 1.6 Hz, 1H), 7.32 (dt, J = 7.7, 1.7 Hz, 2H), 7.07 – 6.99 (m, 2H), 5.89 (s, 2H), 4.10 (A of AB, J_{AB} = 15.2 Hz, 1H), 4.06 (dd, J = 10.2, 6.3 Hz, 1H), 3.88 (B of AB, J_{AB} = 15.1 Hz, 1H), 3.10 – 2.45 (m, 17H), 2.40 (t, J = 7.4 Hz, 2H), 2.12 – 2.00 (m, 1H), 2.02 – 1.90 (m, 1H), 1.93 – 1.79 (m, 1H), 1.75 – 1.58 (m, 3H); ^{13}C NMR (100 MHz, $CDCl_3$) δ: 158.38, 154.56, 147.53, 146.60, 136.68, 134.17, 133.89, 129.94, 121.54, 120.95, 61.76, 57.33, 56.29, 52.81, 51.84, 51.58, 47.00, 44.47, 36.35, 29.27, 28.59, 26.58, 21.91; HRMS (ESI+) calcd for $C_{25}H_{37}N_6$ ($[M+H]^+$): 421.3074. Found: 421.3073, error -0.1 ppm; HPLC/MS purity (> 95%) rt = 0.573 at 254nm, 75-95% MeOH over 3 minutes.

(*S*)-*tert*-butyl-7-(((3-(4-(*tert*-butoxycarbonyl)piperazin-1-yl)propyl)((*S*)-5,6,7,8-tetrahydroquinolin-8-yl)amino)methyl)-7,8-dihydro-1,6-naphthyridine-6(5*H*)-carboxylate (30):

^{1}H NMR (400 MHz, $CDCl_3$) δ: 8.35 (dd, J = 4.9, 1.6 Hz, 1H), 8.27 (dd, J = 4.7, 1.7 Hz, 1H), 7.29 (d, J = 7.4 Hz, 1H), 7.19 (d, J = 7.7 Hz, 1H), 7.04 (dd, J = 7.7, 4.8 Hz, 1H), 6.91 (dd, J = 7.7, 4.7 Hz, 1H), 4.69 (A of AB, J_{AB} = 17.3 Hz, 1H), 4.67 (br s, 0.5H), 4.43 (br s, 0.5H), 4.14 (B of AB, J_{AB} = 17.3 Hz, 1H), 3.96 (dd, J = 9.3, 5.8 Hz, 1H), 3.43–3.31 (m, 5H), 3.27 (A of AB, J_{AB} = 16.7 Hz, 1H), 2.99 (B of ABX, J_{AB} = 16.6, J_{BX} = 6.1 Hz, 1H), 2.95–2.79 (m, 1H), 2.58–2.43 (m, 3H), 2.36–2.09 (m, 7H), 1.97–1.90 (m, 1H), 1.85–1.78 (m, 1H), 1.61–1.36 (m, 4H), 1.43 (s, 9H), 1.42 (s, 9H). LC-MS [(ESI-API, 254 nm) 75-95% MeOH in H_2O (0.1% HCO_2H), 6 min, 1.00 mL/min, C18 (Agilent Zorbax XDB-18, 50 mm x 4.6 mm, 3.5 μm)]: m/z = 621.2 $(M+H)^+$, t = 0.523 min; HPLC/MS purity (> 95%) rt = 0.523 min at 254nm, 75-95% MeOH over 6 minutes.

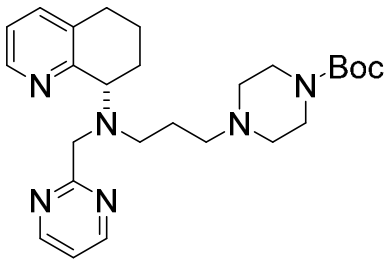
(*S*)-N-(3-(piperazin-1yl)propyl)-N-(((*S*)-5,6,7,8-tetrahydro-1,6-naphthyridin-7-yl)methyl)-5,6,7,8-tetrahydroquinolin-8-amine (31)



General procedure for Boc deprotection starting with **30** (0.057 g, 98% yield, light yellow foam).

^{1}H NMR (400 MHz, $CDCl_3$, ppm) δ: 8.45 (dd, *J* = 4.7, 1.7 Hz, 1H), 8.32 (dd, *J* = 4.8, 1.7 Hz, 1H), 7.34 – 7.28 (m, 2H), 7.01 (dt, *J* = 7.8, 5.1 Hz, 2H), 4.43 (br s, 2H), 4.05 (s, 2H), 4.00 (dd, *J* = 9.7, 5.9 Hz, 1H), 3.03 (tt, *J* = 10.6, 3.4 Hz, 1H), 2.89 (t, *J* = 5.0 Hz, 4H), 2.85 – 2.24 (m, 14H), 2.16 – 2.07 (m, 1H), 2.03 – 1.93 (m, 1H), 1.87 – 1.73 (m, 1H), 1.74 – 1.60 (m, 3H); ^{13}C NMR (100 MHz, $CDCl_3$) δ: 157.76, 154.97, 147.32, 147.15, 136.58, 134.23, 134.11, 130.58, 121.69, 120.84, 60.77, 57.23, 56.61, 53.30, 52.69, 49.88, 47.27, 45.29, 36.60, 29.28, 26.09, 24.19, 21.53; LC-MS [(ESI-API, 254 nm) 75-95% MeOH in H_2O (0.1% HCO_2H), 3 min, 1.00 mL/min, C18 (Agilent Zorbax XDB-18, 50 mm x 4.6 mm, 3.5 μm)]: *m/z* = 421.2 (M + H), 211.2 (M/2 + H), t = 0.490 min.; HRMS (ESI+) calcd for $C_{25}H_{37}N_6$ $(M+H)^+$: 421.3074. Found: 421.3070; HPLC/MS purity (> 95%) rt = 0.490 min at 254nm, 75-95% MeOH over 3 minutes.

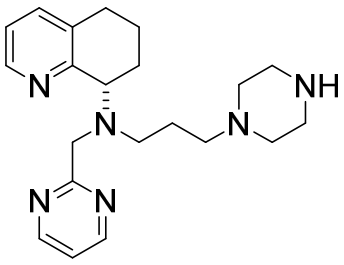
(S)-*tert*-butyl 4-(3-((pyrimidin-2-ylmethyl)(5,6,7,8-tetrahydroquinolin-8-yl)amino)propyl)piperazine-1-carboxylate (32)



Compound **23** (0.387 g, 1.03 mmol), 2-(chloromethyl)pyrimidine hydrochloride (0.359 g, 1.06 mmol), diisopropyl ethylamine (1.08 ml, 6.20 mmol) and potassium iodide (0.017 g, 0.100 mmol) were suspended in 10 mL of acetonitrile and heated at 65^{o}C overnight. The reaction mixture was diluted with EtOAc, washed with saturated aqueous $NaHCO_3$ solution, water and brine, dried over anhydrous $MgSO_4$, filtered and evaporated. The crude product was used without purification for the next step.

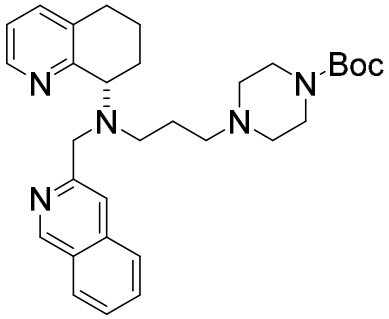
LC-MS [(ESI-API, 254 nm) 75-95% MeOH in H_2O (0.1% HCO_2H), 3 min, 1.00 mL/min, C18 (Agilent Zorbax XDB-18, 50 mm x 4.6 mm, 3.5 µm)]: *m/z* = 467.2 $(M+H)^+$, t = 0.504 min.

(*S*)-N-(3-(piperazin-1yl)propyl)-N-(pyrimidin-2ylmethyl)-5,6,7,8-tetrahydroquinolin-8-amine (33)



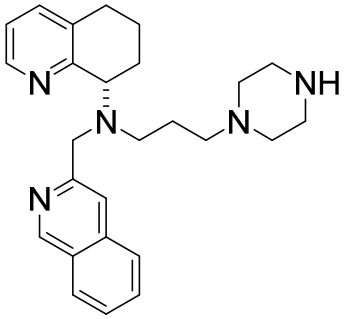
General procedure for Boc deprotection starting with **32** (g, 22% yield over two steps, light yellow foam). ^{1}H NMR (500 MHz, $CDCl_3$) δ 8.79 – 8.71 (m, 4H), 7.26 – 7.23 (t, *J* = 4.9 Hz, 1H), 7.21 (t, *J* = 4.9 Hz, 1H), 4.24 – 4.16 (m, 2H), 4.16 – 4.10 (m, 3H), 3.50 – 3.26 (m, 1H), 3.16 (m, 3H), 3.04 – 2.89 (m, 1H), 2.88 – 2.56 (m, 6H), 2.59 – 2.25 (m, 1H), 2.22 – 1.90 (m, 3H), 1.85 – 1.60 (m, 2H), 1.30 – 1.19 (m, 1H), 0.90 – 0.74 (m, 1H); ^{13}C NMR (151 MHz, $CDCl_3$) δ 168.36, 160.69, 157.08, 146.87, 137.45, 131.81, 121.82, 119.31, 60.62, 56.18, 56.01, 53.48, 51.74, 45.58, 43.59, 29.70, 24.63; LC-MS [(ESI-API, 254 nm) 75-95% MeOH in H_2O (0.1% HCO_2H), 3 min, 1.00 mL/min, C18 (Agilent Zorbax XDB-18, 50 mm x 4.6 mm, 3.5 µm)]: *m/z* = 367.2 $(M + H)^+$, t = 0.504 min.; HRMS Calc. for $C_{21}H_{31}N_6$ $(M+H)^+$:367.26047, Found: 367.26092; HPLC/MS purity (> 95%) rt = 0.550 min at 254nm, 75-95% MeOH over 3 minutes.

(*S*)-*tert*-butyl 4-(3-((isoquinolin-3-ylmethyl)(5,6,7,8-tetrahydroquinolin-8-yl)amino)propyl)piperazine-1-carboxylate (34)



To the solution of compound **23** (0.300 g, 0.801mmol) in 10 mL DCE, isoquinoline-3-carbaldehyde (0.126 g, 0.801 mmol) and sodium triacetoxyhydroborate (0.340 g, 1.60 mmol) were added and the suspension was stirred at room temperature for overnight. The reaction mixture was quenched by addition of saturated aqueous $NaHCO_3$ solution. The aqueous phase was extracted with DCM; the combined organic layers were washed with water, dried over anhydrous $MgSO_4$, filtered and evaporated to give compound **34**. The crude product was used without purification for the next step.

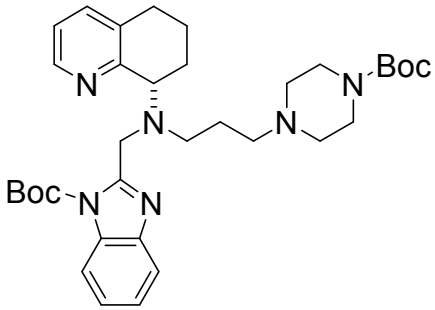
(*S*)-N-(isoquinolin-3-ylmethyl)-N-(3-piperazin-1yl)propyl)-5,6,7,8-tetrahydroquinolin-8- amine (35)



General procedure for Boc deprotection starting with **34** (0.108 g, 52% yield over two steps, light yellow foam). ^{1}H NMR (500 MHz, $CDCl_3$) δ 9.19 (s, 1H), 8.50 (d, *J* = 4.8 Hz, 1H), 8.01 (s, 1H), 7.95 (d, *J* = 8.3 Hz, 1H), 7.84 (d, *J* = 8.5 Hz, 1H), 7.68 (ddd, *J* = 8.2, 6.9, 1.2 Hz, 1H), 7.56 (ddd, *J* = 8.1, 6.9, 1.1 Hz, 1H), 7.36 (d, *J* = 7.5 Hz, 1H), 7.06 (ddd, *J* = 7.7, 4.7, 0.7 Hz, 1H), 4.24 – 4.09 (m, 2H), 4.02 (d, *J* = 15.1 Hz, 1H), 3.11 – 2.97 (m, 4H), 2.93 – 2.76 (m, 3H), 2.75 – 2.57 (m, 6H), 2.53 – 2.44 (m, 2H), 2.30 – 2.18 (m, 1H), 2.10 – 1.99 (m, 1H), 1.98 – 1.87 (m, 1H), 1.81 – 1.54 (m, 3H); ^{13}C NMR (126 MHz, $CDCl_3$) δ 151.89, 147.07, 136.74, 136.44, 134.44, 130.42, 127.67, 127.53, 126.77, 126.54, 121.76, 118.93, 61.21, 57.68, 56.41, 50.75, 50.56, 43.60, 29.24, 21.48; LC-MS [(ESI-API,

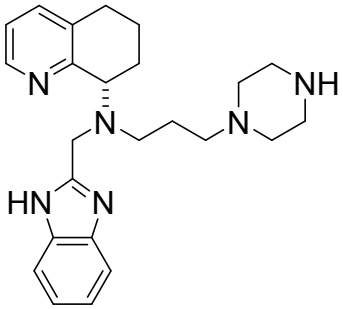
254 nm) 75-95% MeOH in H_2O (0.1% HCO_2H), 3 min, 1.00 mL/min, C18 (Agilent Zorbax XDB-18, 50 mm x 4.6 mm, 3.5 µm)]: *m/z* = 416.2 $(M+H)^+$, t = 0.897 min.; HRMS Calc. for $C_{26}H_{34}N_5$ $(M+H)^+$: 416.28087; Found: 416.28096; HPLC/MS purity (> 95%) rt = 0.590 min at 254nm, 75-95% MeOH over 3 minutes.

(*S*)-*tert*-butyl 2-(((3-(4-(tert-butoxycarbonyl)piperazin-1-yl)propyl)(5,6,7,8-tetrahydroquinolin-8-yl)amino)methyl)-1H-benzo[d]imidazole-1-carboxylate (36)



Compound **23** (0.5 g, 1.34 mmol), tert-butyl 2-(chloromethyl)-1H-benzo[d]imidazole-1-carboxylate (0.534 g, 2.00 mmol), diisopropylethylamine (0.93 ml, 5.34 mmol) and potassium iodide (0.0220 g, 0.130 mmol) were suspended in 20 mL of acetonitrile and the mixture was heated at 65°C overnight. The reaction mixture was diluted with EtOAc, washed with saturated aqueous $NaHCO_3$ solution, water and brine, dried over anhydrous $MgSO_4$, filtered and evaporated. The crude product was used without purification for the next step.

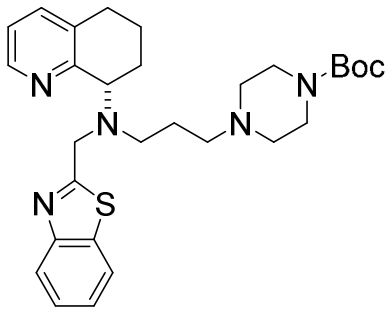
(*S*)-N-((1H-benzo[*d*]imidazole-2yl)methyl)-N-(3-(piperazine-1yl)propyl)-5,6,7,8-tetrahydroquinolin-8-amine (37)



General procedure for Boc deprotection starting with **36** (0.077 g, 38% yield over two steps, yellow foam).

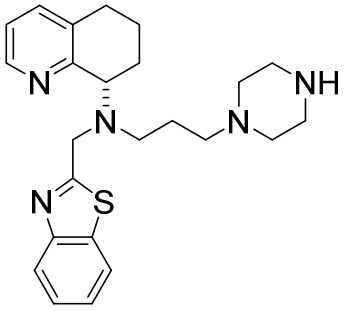
^{1}H NMR (500 MHz, $CDCl_3$) δ 8.57 (dd, *J* = 4.8, 1.7 Hz, 1H), 7.58 (dt, *J* = 7.1, 3.6 Hz, 2H), 7.47 – 7.44 (m, 1H), 7.22 – 7.15 (m, 3H), 4.15 – 4.09 (m, 1H), 4.09 – 4.03 (m, 2H), 2.97 – 2.93 (m, 3H), 2.88 – 2.81 (m, 1H), 2.75 – 2.69 (m, 2H), 2.59 (dt, *J* = 12.9, 7.5 Hz, 1H), 2.53 – 2.35 (m, 6H), 2.31 – 2.18 (m, 2H), 2.09 – 1.99 (m, 1H), 1.97 – 1.84 (m, 1H), 1.77 – 1.63 (m, 2H), 1.62 – 1.50 (m, 2H), 1.29 – 1.23 (m, 1H); LC-MS (ESI-API, 254 nm) 75-95% MeOH in H_2O (0.1% HCO_2H), 3 min, 1.00 mL/min, C18 (Agilent Zorbax XDB-18, 50 mm x 4.6 mm, 3.5 μm), *m/z* = 405.2 (M + H) t = 0.513 min; HRMS Calc. for $C_{24}H_{33}N_6$ (M+H):405.27612, Found: 405.27644; HPLC/MS purity (> 95%) rt = 0.584 min at 254nm, 75-95% MeOH over 3 minutes.

(*S*)-*tert*-butyl 4-(3-((benzo[d]thiazol-2-ylmethyl)(5,6,7,8-tetrahydroquinolin-8-yl)amino)propyl)piperazine-1-carboxylate (38)



Compound **23** (0.387 g, 1.03 mmol), 2-(chloromethyl)benzo[d]thiazole (0.300 g, 1.06 mmol), diisopropylethylamine (0.72 ml, 4.13 mmol) and potassium iodide (0.0170 g, 0.1 mmol) were suspended in 10 mL of acetonitrile and the suspension was heated at 65^{o}C for overnight. The reaction mixture was diluted with EtOAc, washed with saturated aqueous $NaHCO_3$ solution, water and brine, dried over anhydrous $MgSO_4$, filtered and evaporated. The crude product was used without purification in the next step

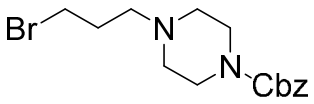
(*S*)-N-((1H-benzo[*d*]thiazol-2yl)methyl)-N-(3-(piperazine-1yl)propyl)-5,6,7,8-tetrahydroquinolin-8-amine (39)



General procedure for Boc deprotection starting with **38** (0.089 g, 42% yield over two steps, yellow foam).

^{1}H NMR (500 MHz, $CDCl_3$) δ 8.46 (dd, *J* = 4.7, 1.8 Hz, 1H), 7.91 (dt, *J* = 8.1, 0.9 Hz, 1H), 7.87 (dt, *J* = 7.9, 0.9 Hz, 1H), 7.43 (ddd, *J* = 8.2, 7.2, 1.3 Hz, 1H), 7.37 – 7.31 (m, 2H), 7.06 (dd, *J* = 7.7, 4.6 Hz, 1H), 4.81 – 4.58 (m, 4H), 4.52 (d, *J* = 16.8 Hz, 1H), 4.19 – 4.13 (m, 1H), 4.04 (d, *J* = 16.7 Hz, 1H), 3.04 – 2.95 (m, 5H), 2.61 – 2.52 (m, 4H), 2.48 (t, *J* = 7.4 Hz, 2H), 2.29 – 2.18 (m, 1H), 2.04 – 1.97 (m, 1H), 1.86 (dddd, *J* = 13.0, 11.8, 9.9, 3.0 Hz, 1H), 1.77 – 1.66 (m, 3H); ^{13}C NMR (126 MHz, $CDCl_3$) δ 177.39, 157.77, 153.63, 146.95, 136.54, 135.31, 134.05, 125.63, 124.48, 122.45, 121.66, 61.33, 56.23, 55.38, 51.81, 50.82, 44.46, 29.23, 28.74, 25.81, 21.52; LC-MS (ESI-API, 254 nm) 75-95% MeOH in H_2O (0.1% HCO_2H), 3 min, 1.00 mL/min, C18 (Agilent Zorbax XDB-18, 50 mm x 4.6 mm, 3.5 µm), *m/z* = 422.2 (M + H) t = 0.515 min; HRMS Calc. for $C_{24}H_{32}N_5S$ (M+H):422.23729, Found: 422.23720; HPLC/MS purity (> 95%) rt = 0.767 min at 254nm, 75-95% MeOH over 3 minutes.

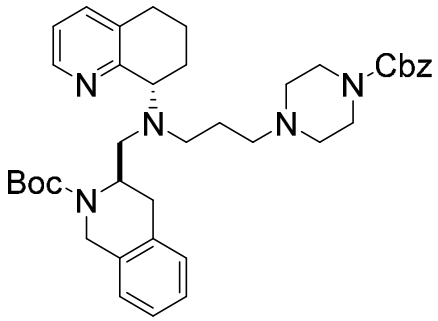
Benzyl 4-(3-bromopropyl)piperazine-1-carboxylate (40):



Benzyl piperazine-1-carboxylate (4.38 ml, 22.0 mmol) was dissolved in dioxane (100 ml) and potassium carbonate (15.2 g, 110 mmol) and 1,3-dibromopropane (22.4 ml, 220 mmol) were added and to the suspension was heated at 65°C for overnight. The reaction mixture was cooled to room temperature, the inorganic by-products were removed via filtration and the filtrate was evaporated. The residue was purified by silica gel column chromatography EtOAc in hexanes as eluent (g, 75% yield, light yellow oil).

^{1}H NMR (400 MHz, $CDCl_3$) δ 7.43 - 7.29 (m, 5H), 5.13 (s, 2H), 3.59 - 3.41 (m, 7H), 2.49 (dd, *J* = 7.3, 6.6 Hz, 2H), 2.46 - 2.34 (m, 5H), 2.07 - 1.95 (m, 2H).

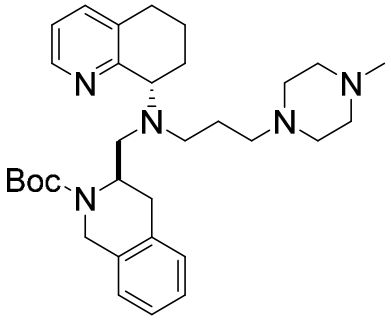
***tert*-butyl-(*R*)-3-(((3-(4-(benzyloxycarbonyl)piperazin-1-yl)propyl)((*S*)-5,6,7,8-tetrahydroquinolin-8-yl)amino)methyl)-3,4-dihydroisoquinoline- 2(1H)-carboxylate (41):**



(*R*)-*tert*-butyl 3-((((*S*)-5,6,7,8-tetrahydroquinolin-8-yl)amino)methyl)-3,4-dihydroisoquinoline-2(1H)-carboxylate (**6**) (2.00 g, 5.08 mmol), diisopropylethylamine (1.78 ml, 10.2 mmol), potassium iodide (0.0840 g, 0.508 mmol), and benzyl 4-(3-bromopropyl)piperazine-1-carboxylate (**40**) (2.60 g, 7.62 mmol) dissolved in 50 ml of acetonitrile were heated at 65°C for overnight. The reaction mixture was cooled to room temperature and saturated aqueous $NaHCO_3$ was added to the solution and the aqueous phase was extracted with DCM. The combined organic layer was dried over anhydrous $MgSO_4$; filtered and evaporated. The residue was purified by silica gel column chromatography using DCM: MeOH: NH_4OH (9:1:0.1) as eluent (2.59 g, 78% yield, off white foam).

^{1}H NMR (400 MHz, $CDCl_3$) δ 7.42 - 7.30 (m, 5H), 7.28 - 7.23 (m, 2H), 7.23 - 6.92 (m, 5H), 5.13 (s, 2H), 4.72 - 4.55 (m, 1H), 4.51 - 4.24 (m, 1H), 4.16 (d, *J* = 17.3 Hz, 1H), 3.62 - 3.41 (m, 6H), 3.14 - 2.87 (m, 2H), 2.87 - 2.53 (m, 3H), 2.55 - 2.21 (m, 6H), 2.08 - 1.85 (m, 5H), 1.50 (s, 9H); LC-MS [(ESI-API, 254 nm) 75-95% MeOH in H2O (0.1% HCO2H), 6 min, 1.00 mL/min, C18 (Agilent Zorbax XDB-18, 50 mm x 4.6 mm, 3.5 µm)]: m/z = 654.4 $(M+H)^+$, t = 3.798 min.

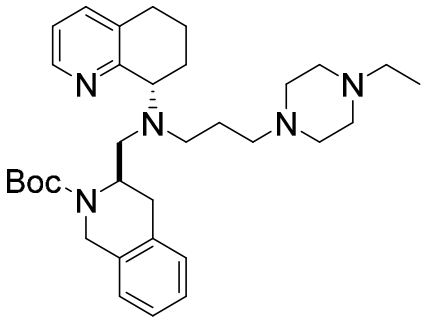
***tert*-butyl-(*R*)-3-(((3-(4-methylpiperazin-1-yl)propyl)((*S*)-5,6,7,8-tetrahydroquinolin-8-yl)amino)methyl)-3,4-dihydroisoquinoline- 2(1H)-carboxylate (42a):**



Step 1: (*R*)-tert-butyl 3-(((3-(4-((benzyloxy)carbonyl)piperazin-1-yl)propyl)((*S*)-5,6,7,8-tetrahydroquinolin-8-yl)amino)methyl)-3,4-dihydroisoquinoline-2(1H)-carboxylate (0.150 g, 0.229 mmol) was dissolved in EtOH and ammonium formate (0.0290 g, 0.459 mmol) and $PdOH_2$ (8.05 mg, 0.0110 mmol) were added and the suspension was heated at reflux for 3 hours. The reaction mixture was cooled to room temperature and filtered through a celite plug and the filtrate was evaporated. The crude product was used without further purification.

Step 2: To the solution of *tert*-butyl-(*R*)-3-(((3-(piperazin-1-yl)propyl)((*S*)-5,6,7,8-tetrahydroquinolin-8-yl)amino)methyl)-3,4-dihydroisoquinoline- 2(1H)-carboxylate (0.215 g, 0.414mmol) in 5 ml DCE, paraformaldehyde (0.124 g, 4.14 mmol) and sodium triacetoxyhydroborate (0.181 g, 0.827 mmol) were added and the suspension was stirred at room temperature for overnight. The reaction mixture was quenched by addition of saturated aqueous $NaHCO_3$ solution. The aqueous phase was extracted with DCM; the combined organic layers were washed with water, dried over anhydrous $MgSO_4$, filtered and evaporated. The crude product was used without purification for the next step.

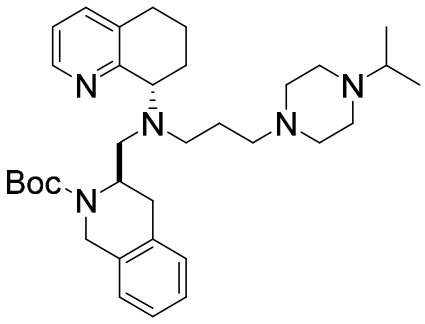
tert-butyl-(_R_)-3-(((3-(4-ethylpiperazin-1-yl)propyl)((_S_)-5,6,7,8-tetrahydroquinolin-8-yl)amino)methyl)-3,4-dihydroisoquinoline- 2(1H)-carboxylate (42b):



Step 1: (*R*)-tert-butyl 3-(((3-(4-((benzyloxy)carbonyl)piperazin-1-yl)propyl)((*S*)-5,6,7,8-tetrahydroquinolin-8-yl)amino)methyl)-3,4-dihydroisoquinoline-2(1H)-carboxylate (0.150 g, 0.229 mmol) was dissolved in EtOH and ammonium formate (0.0290 g, 0.459 mmol) and $Pd(OH)_2$ (8.05 mg, 0.0110 mmol) were added. The suspension was heated at reflux for 3 hours. The reaction mixture was cooled to room temperature and filtered through celite plug and the filtrate was evaporated. The crude product was used without further purification.

Step 2: To the solution of *tert*-butyl-(*R*)-3-(((3-(piperazin-1-yl)propyl)((*S*)-5,6,7,8-tetrahydroquinolin-8-yl)amino)methyl)-3,4-dihydroisoquinoline- 2(1H)-carboxylate (0.215 g, 0.414 mmol) in 5 ml DCE, acetaldehyde (0.12 ml, 2.07 mmol) and sodium triacetoxyhydroborate (0.181 g, 0.827 mmol) were added and the suspension was stirred at room temperature for overnight. The reaction mixture was quenched by addition of saturated aqueous $NaHCO_3$ solution. The aqueous phase was extracted with DCM; combined organic layers were washed with water, dried over anhydrous $MgSO_4$, filtered and evaporated. The crude product was used without purification for the next step.

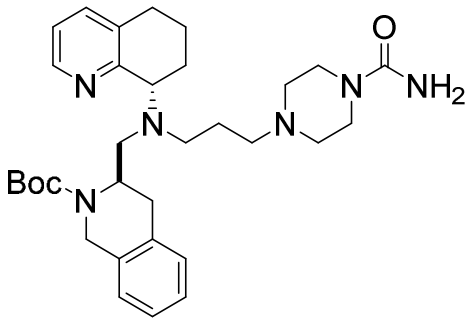
tert-butyl-(_R_)-3-(((3-(4-isopropylpiperazin-1-yl)propyl)((_S_)-5,6,7,8-tetrahydroquinolin-8-yl)amino)methyl)-3,4-dihydroisoquinoline- 2(1H)-carboxylate (42c):



Step 1: (*R*)-tert-butyl 3-(((3-(4-((benzyloxy)carbonyl)piperazin-1-yl)propyl)((*S*)-5,6,7,8-tetrahydroquinolin-8-yl)amino)methyl)-3,4-dihydroisoquinoline-2(1H)-carboxylate (0.150 g, 0.229 mmol) was dissolved in EtOH (10 ml), ammonium formate (0.0290 g, 0.459 mmol) and $Pd(OH)_2$ (8.05 mg, 0.0110 mmol) were added and the suspension was heated at reflux for 3 hours. The reaction mixture was cooled to room temperature and filtered through a celite plug and the filtrate was evaporated.The crude product was used in next step without further purification.

Step 2: To the solution of *tert*-butyl-(*R*)-3-(((3-(piperazin-1-yl)propyl)((*S*)-5,6,7,8-tetrahydroquinolin-8-yl)amino)methyl)-3,4-dihydroisoquinoline- 2(1H)-carboxylate (0.193 g, 0.414 mmol) in 3 mL acetone, sodium triacetoxyhydroborate (0.203 g, 0.928 mmol) was added and the suspension was stirred at room temperature for overnight. The reaction mixture was quenched by addition of saturated aqueous $NaHCO_3$ solution. The aqueous phase was extracted with DCM; the combined organic layers were washed with water, dried over anhydrous $MgSO_4$, filtered and evaporated. The crude product was used without purification for the next step.

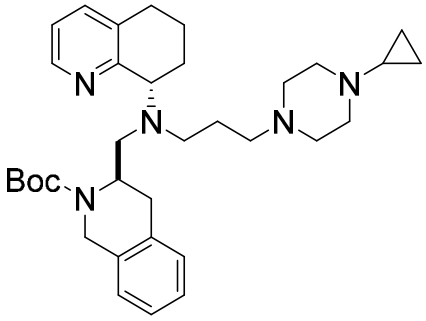
tert-butyl-(_R_)-3-(((3-(4-carbomoylpiperazin-1-yl)propyl)((_S_)-5,6,7,8-tetrahydroquinolin-8-yl)amino)methyl)-3,4-dihydroisoquinoline- 2(1H)-carboxylate (42d):



Step 1: (*R*)-tert-butyl 3-(((3-(4-((benzyloxy)carbonyl)piperazin-1-yl)propyl)((*S*)-5,6,7,8-tetrahydroquinolin-8-yl)amino)methyl)-3,4-dihydroisoquinoline-2(1H)-carboxylate (0.150 g, 0.229 mmol) was dissolved in EtOH (10 ml), ammonium formate (0.0290 g, 0.459 mmol) and $Pd(OH)_2$ (8.05 mg, 0.011 mmol) were added. The suspension was heated at reflux for 3 hours. The reaction mixture was cooled to room temperature and filtered through a celite plug and the filtrate was evaporated. The crude product was used in next step without further purification.

Step 2: (*R*)-tert-butyl 3-(((3-(piperazin-1-yl)propyl)((*S*)-5,6,7,8-tetrahydroquinolin-8-yl)amino)methyl)-3,4-dihydroisoquinoline-2(1H)-carboxylate (0.215 g, 0.414 mmol) was dissolved in 5 ml THF and DIPEA (0.16 ml, 0.910 mmol) and isocyanatotrimethylsilane (0.07 ml, 0.463 mmol) were added dropwise to the reaction mixture. The solution was stirred at room temperature for overnight. The reaction mixture was poured into water and extracted with DCM (2x). The combined organic layers were dried over anhydrous $MgSO_4$, filtered and evaporated and used for the next step.

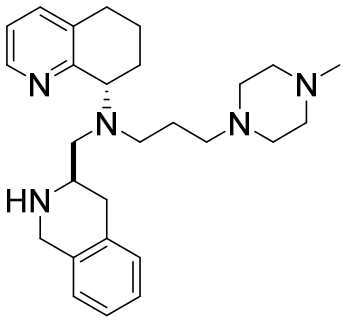
tert-butyl-(_R_)-3-(((3-(4-cyclopropylpiperazin-1-yl)propyl)((_S_)-5,6,7,8-tetrahydroquinolin-8-yl)amino)methyl)-3,4-dihydroisoquinoline- 2(1H)-carboxylate (42e):



Step 1: (*R*)-tert-butyl 3-(((3-(4-((benzyloxy)carbonyl)piperazin-1-yl)propyl)((*S*)-5,6,7,8-tetrahydroquinolin-8-yl)amino)methyl)-3,4-dihydroisoquinoline-2(1H)-carboxylate (0.150 g, 0.229 mmol) was dissolved in EtOH (10 ml) ,ammonium formate (0.0290 g, 0.459 mmol) and $Pd(OH)_2$ (8.05 mg, 0.0110 mmol) were added. The suspension was heated at reflux for 3 hours. The reaction mixture was cooled to room temperature and filtered through a celite plug and the filtrate was evaporated. The crude product was used in the next step without further purification.

Step 2: To the solution of *tert*-butyl-(*R*)-3-(((3-(piperazin-1-yl)propyl)((*S*)-5,6,7,8-tetrahydroquinolin-8-yl)amino)methyl)-3,4-dihydroisoquinoline- 2(1H)-carboxylate (0.193 g, 0.414mmol) in 4 ml MeOH, (1-ethoxycyclopropoxy)trimethylsilane (0.45 ml, 2.23 mmol), acetic acid (0.21 ml, 3..71 mmol) and sodium cyanoborohydride (0.117 g, 1.86 mmol) were added and the suspension was stirred at room temperature for overnight. The reaction mixture was quenched by addition of saturated aqueous $NaHCO_3$ solution and extracted with DCM; the combined organic layers were washed with water and dried over anhydrous $MgSO_4$,filtered and evaporated. The crude product was used without purification for the next step.

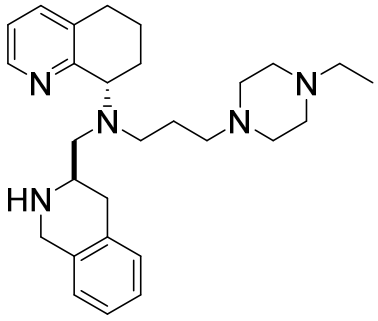
(*S*)-N-(3-(4-methylpiperazin-1-yl)propyl)-N-(((*R*)-1,2,3,4-tetrahydroisoquinolin-3-yl)methyl)-5,6,7,8-tetrahydroquinolin-8-amine (43):



General procedure for Boc deprotection starting with **42a** (0.102 g, 47% yield over three steps, off-white foam).

^{1}H NMR (400 MHz, $CDCl_3$) δ 8.41 (dd, *J* = 4.8, 1.7 Hz, 1H), 7.31 (dd, *J* = 8.0, 3.2 Hz, 1H), 7.08 – 6.99 (m, 5H), 4.06 (dd, *J* = 15.5, 8.2 Hz, 2H), 3.90 (d, *J* = 15.0 Hz, 1H), 3.22 – 3.00 (m, 4H), 2.94 (dd, *J* = 13.3, 3.2 Hz, 1H), 2.83 – 2.69 (m, 3H), 2.63 – 2.55 (m, 2H), 2.50 – 2.31 (m, 6H), 2.26 (s, 3H), 2.10 – 2.01 (m, 1H), 2.00 – 1.91 (m, 1H), 1.91 – 1.81 (m, 1H), 1.75 – 1.59 (m, 5H), 1.32 – 1.21 (m, 2H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 158.86, 146.82, 136.69, 135.37, 134.68, 134.13, 129.24, 126.59, 126.14, 125.70, 121.57, 61.55, 57.81, 56.71, 55.27, 53.44, 52.57, 52.38, 48.58, 46.19, 33.70, 29.57, 29.19, 27.14, 22.13; LC-MS (ESI-API, 254 nm) 75-95% MeOH in H_2O (0.1% HCO_2H), 3 min, 1.00 mL/min, C18 (Agilent Zorbax XDB-18, 50 mm x 4.6 mm, 3.5 µm), *m/z* = 434.2 (M + H) t = 1.355 min; HRMS Calc. for $C_{27}H_{40}N_5$ (M+H): 434.32782, Found: 434.32778; HPLC/MS purity (> 95%) rt = 1.767 min at 254nm, 75-95% MeOH over 3 minutes.

(*S*)-N-(3-(4-ethylpiperazin-1-yl)propyl)-N-(((*R*)-1,2,3,4-tetrahydroisoquinolin-3-yl)methyl)-5,6,7,8-tetrahydroquinolin-8-amine (44):

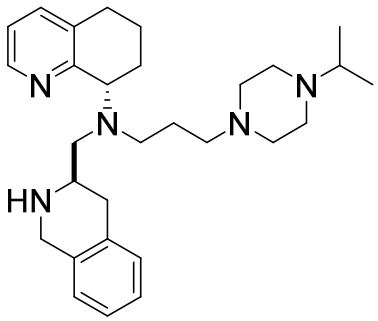


General procedure for Boc deprotection starting with **42b** (0.136 g, 61% yield over three steps, off white foam).

^{1}H NMR (400 MHz, $CDCl_3$) δ 8.39 (dd, *J* = 4.7, 1.7 Hz, 1H), 7.29 (dd, *J* = 7.7, 1.7 Hz, 1H), 7.07 – 6.94 (m, 5H), 4.08 – 4.04 (m, 1H), 4.01 (d, *J* = 15.1 Hz, 1H), 3.86 (d, *J* = 15.1 Hz, 1H), 3.45 (q, *J* = 7.0 Hz, 1H), 3.03 (ddd, *J* = 13.3, 7.7, 5.8 Hz, 1H), 2.93 (dd, *J* = 13.2, 3.2 Hz, 1H), 2.86 – 2.68 (m, 6H), 2.69 – 2.50 (m, 4H), 2.48 – 2.15 (m, 8H), 2.09 – 1.99 (m, 1H), 1.99 – 1.90 (m, 1H), 1.85 (ddd, *J* = 12.7, 9.9, 2.7 Hz, 1H), 1.68 (tdd, *J* = 14.3, 11.6, 7.4 Hz, 4H), 1.04 (t, *J* = 7.2 Hz, 3H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 158.81, 146.84, 136.67, 135.60, 134.76, 134.12, 129.23, 126.59, 126.08, 125.65, 121.56, 66.00, 61.51, 57.90, 56.77, 53.43, 52.91, 52.53, 52.45, 48.72, 33.84, 29.59, 29.04, 27.12, 22.13, 12.15; LC-MS (ESI-API, 254 nm) 75-95% MeOH in H_2O (0.1% HCO_2H), 3 min, 1.00 mL/min, C18 (Agilent Zorbax XDB-18, 50 mm x 4.6 mm, 3.5 µm), *m/z* = 448.6 (M + H) t = 1.051 min; HRMS Calc. for

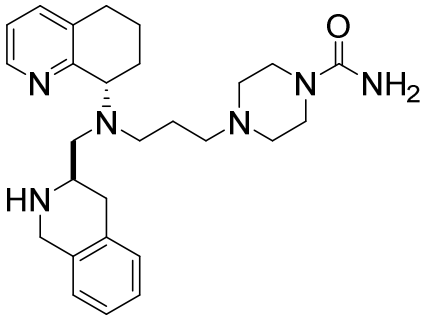
$C_{28}H_{42}N_5$ (M+H): 448.34347, Found: 448.34539; HPLC/MS purity (> 95%) rt = 1.105 min at 254nm, 75-95% MeOH over 3 minutes.

(*S*)-N-(3-(4-isopropylpiperazin-1-yl)propyl)-N-(((*R*)-1,2,3,4-tetrahydroisoquinolin-3-yl)methyl)-5,6,7,8-tetrahydroquinolin-8-amine (45):



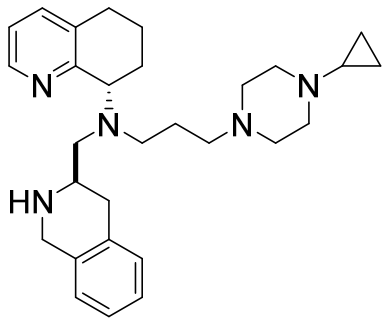
General procedure for Boc deprotection starting with **42c** (0.104 g, 45% yield over three steps, off-white foam). ^{1}H NMR (500 MHz, $CDCl_3$) δ 8.41 (dd, *J* = 4.8, 1.7 Hz, 1H), 7.30 (dd, *J* = 7.7, 2.0 Hz, 1H), 7.09 – 6.98 (m, 5H), 4.10 – 4.00 (m, 2H), 3.91 (d, *J* = 14.9 Hz, 1H), 3.13 (ddd, *J* = 13.2, 7.7, 5.5 Hz, 1H), 2.91 (dd, *J* = 13.2, 3.3 Hz, 1H), 2.81 – 2.70 (m, 2H), 2.63 (dddd, *J* = 13.1, 10.9, 6.0, 2.5 Hz, 4H), 2.55 – 2.41 (m, 13H), 2.10 – 2.01 (m, 1H), 1.95 (dddd, *J* = 15.0, 13.1, 6.6, 4.0 Hz, 1H), 1.92 – 1.81 (m, 1H), 1.77 – 1.64 (m, 3H), 1.03 (d, *J* = 6.6 Hz, 6H); ^{13}C NMR (126 MHz, $CDCl_3$) δ 158.96, 146.84, 136.61, 134.83, 134.11, 131.15, 129.24, 126.59, 126.04, 125.62, 121.52, 61.34, 57.85, 56.88, 54.59, 53.83, 53.71, 52.67, 52.52, 48.86, 33.89, 29.58, 29.47, 27.18, 22.15, 18.86; LC-MS (ESI-API, 254 nm) 75-95% MeOH in H_2O (0.1% HCO_2H), 3 min, 1.00 mL/min, C18 (Agilent Zorbax XDB-18, 50 mm x 4.6 mm, 3.5 µm), *m/z* = 462.3 (M + H) t = 0.893 min; HRMS Calc. for $C_{29}H_{44}N_5$ (M+H): 462.35912, Found: 462.36188; HPLC/MS purity (> 95%) rt = 0.901 min at 254nm, 75-95% MeOH over 3 minutes.

4-3-(((*R*)-1,2,3,4-tetrahydroisoquinolin-3-yl)methyl)((*S*)-5,6,7,8-tetrahydroquinolin-8-yl)amino)propyl)piperazin-1-carboxamide (46):



General procedure for Boc deprotection starting with **42d** (0.093 g, 40% yield over three steps, white foam). ^{1}H NMR (400 MHz, CD_3OD) δ 8.50 (dd, *J* = 4.9, 1.6 Hz, 1H), 7.63 (dd, *J* = 7.9, 1.6 Hz, 1H), 7.32 - 7.19 (m, 7H), 4.49 (d, J = 6.1 Hz, 2H), 4.14 (dd, *J* = 10.5, 6.1 Hz, 1H), 3.65 - 3.61 (m, 5H), 3.32 - 3.25 (m, 2H), 3.16-3.07 (m, 6H), 3.07 - 2.93 (m, 3H), 2.91-2.79 (m, 2H), 2.76 - 2.56 (m, 3H), 2.25 - 2.16 (m, 1H), 2.11-2.00 (m, 1H), 1.81 - 1.71 (m, 1H), 1.60-1.53 (brs, 1H), 1.35 (d, J = 6.6 Hz, 1H); ^{13}C NMR (100 MHz, CD_3OD) δ 158.87, 156.42, 145.80, 138.81, 135.60, 130.99, 128.76, 128.09, 127.73, 126.75, 126.31, 122.69, 64.20, 55.83, 54.74, 53.37, 51.30, 44.17, 40.86, 29.02, 28.48, 24.86, 23.46, 21.30; LC-MS (ESI-API, 254 nm) 75-95% MeOH in H_2O (0.1% HCO_2H), 3 min, 1.00 mL/min, C18 (Agilent Zorbax XDB-18, 50 mm x 4.6 mm, 3.5 μm), *m/z* = 463.5 (M + H) t = 0.886 min; HRMS Calc. for $C_{27}H_{39}N_5O$ (M+H): 463.31799, Found: 463.31809; HPLC/MS purity (> 95%) rt = 0.995 min at 254nm, 75-95% MeOH over 3 minutes.

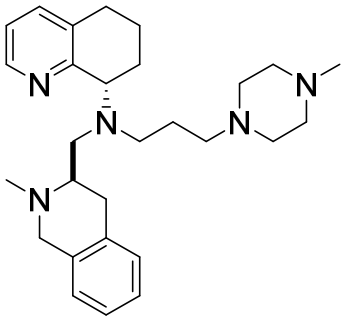
(*S*)-N-(3-(4-cyclopropylpiperazin-1-yl)propyl)-N-(((*R*)-1,2,3,4-tetrahydroisoquinolin-3-yl)methyl)-5,6,7,8-tetrahydroquinolin-8-amine (47):



General procedure for Boc deprotection starting with **42e** (0.108 g, 47% over three steps, light yellow foam).

^{1}H NMR (500 MHz, $CDCl_3$) δ 8.41 (d, *J* = 4.7 Hz, 1H), 7.30 (d, *J* = 7.7 Hz, 1H), 7.09 - 7.05 (m, 2H), 7.04 - 6.98 (m, 3H), 4.11 - 4.00 (m, 3H), 3.91 (d, *J* = 14.9 Hz, 1H), 3.17 - 3.07 (m, 1H), 2.92 (dd, *J* = 13.3, 3.2 Hz, 1H), 2.81 - 2.71 (m, 3H), 2.67 - 2.58 (m, 6H), 2.47 - 2.30 (m, 6H), 2.10 - 2.00 (m, 1H), 2.00 - 1.92 (m, 1H), 1.91 - 1.82 (m, 1H), 1.75 - 1.62 (m, 5H), 1.61 - 1.55 (m, 1H), 0.43 - 0.41 (m, 2H), 0.38 - 0.36 (m, 2H); ^{13}C NMR (126 MHz, $CDCl_3$) δ 158.94, 146.83, 136.62, 134.80, 134.11, 131.16, 129.24, 126.59, 126.06, 125.62, 121.52, 61.36, 57.84, 56.84, 53.48, 53.40, 53.31, 52.51, 48.80, 38.63, 33.86, 29.57, 29.45, 27.19, 22.15, 5.84; LC-MS (ESI-API, 254 nm) 75-95% MeOH in H_2O (0.1% HCO_2H), 3 min, 1.00 mL/min, C18 (Agilent Zorbax XDB-18, 50 mm x 4.6 mm, 3.5 µm), *m/z* = 460.3 (M + H) t = 1.210 min; HRMS Calc. for $C_{29}H_{42}N_5$ (M+H): 460.34347, Found: 460.34350; HPLC/MS purity (> 95%) rt = 1.308 min at 254nm, 75-95% MeOH over 3 minutes.

(*S*)-N-(((*S*)-2-methyl-1,2,3,4-tetrahydroisoquinolin-3-yl)methyl)-N-(3-(4-methylpiperazin-1-yl)propyl)-5,6,7,8-tetrahydroquinolin-8-amine (48):



The amine **17** (0.100 g, 0.238 mmol) was dissolved in DCM; paraformaldehyde (0.0720 g, 2.38 mmol) and sodium triacetoxyborohydride (0.156 g, 0.715 mmol) were added and the suspension was stirred for overnight. The reaction mixture was quenched by addition of saturated aqueous $NaHCO_3$ solution. The organic phase was dried over anhydrous $MgSO_4$, filtered and evaporated. The residue was purified by silica gel column chromatography using DCM: MeOH: NH_3 (8:2:0.6) as eluent (0.021 g, 20% yield, off-white foam).

^{1}H NMR (500 MHz, $CDCl_3$) δ 8.48 (d, *J* = 4.5 Hz, 1H), 7.39 (d, *J* = 7.8 Hz, 1H), 7.23 - 7.09 (m, 5H), 4.43 - 4.25 (m, 2H), 4.03 - 3.80 (m, 2H), 3.21 (dd, *J* = 16.3, 7.4 Hz, 1H), 2.92 (d, *J* = 3.9 Hz, 3H), 2.78 - 2.51 (m, 14H), 2.38 (d, *J* = 4.0 Hz, 3H), 2.20 - 2.05 (m, 2H), 1.98 (dt, *J* = 18.9, 7.2 Hz, 2H), 1.86 - 1.65 (m, 2H), 1.65 - 1.51 (m, 3H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 153.36, 146.69, 138.41, 134.20, 133.20, 128.90, 128.73, 127.60, 126.73, 122.71, 122.50, 59.29, 55.23, 53.53, 53.06, 52.83, 51.54, 51.33, 44.78, 29.76, 29.25, 28.53, 25.00, 21.41; LC-MS (ESI-API, 254 nm)

75-95% MeOH in H_2O (0.1% HCO_2H), 3 min, 1.00 mL/min, C18 (Agilent Zorbax XDB-18, 50 mm x 4.6 mm, 3.5 µm), *m/z* = 448.2 (M + H) t = 0.922 min; HRMS Calc. for $C_{28}H_{42}N_5$ (M+H): 448.34347, Found: 448.34359; HPLC/MS purity (> 95%) rt = 0.925 min at 254nm, 75-95% MeOH over 3 minutes.

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