Supporting Information

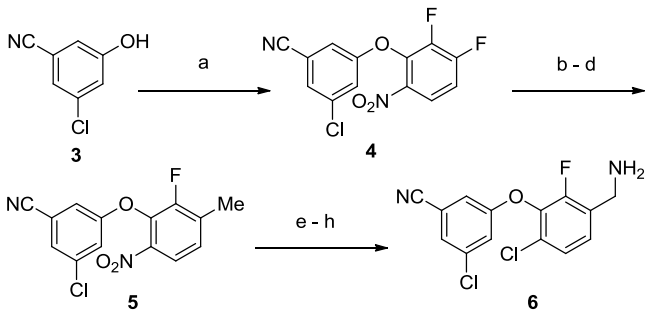
Rational Design of Potent Non-Nucleoside Inhibitors of HIV Reverse Transcriptase

Pek Chong, Paul Sebahar[†], Michael Youngman, Dulce Garrido, Huichang Zhang, Eugene Stewart, Robert T. Nolte, Liping Wang, Robert G. Ferris, Mark Edelstein, Kurt Weaver, Amanda Mathis and Andrew Peat*

GlaxoSmithKline Research & Development, 5 Moore Drive, Research Triangle Park, NC 27709, USA

Methods Described:

I. Experimental procedures



Compound 4. **3-chloro-5-[(2,3-difluoro-6-nitrophenyl)oxy]benzonitrile.** 3-Chloro-5-hydroxybenzonitrile (20.0 g, 130 mmol) was dissolved in anhydrous THF (500 mL) and cooled to 0 °C. Sodium hydride (60% dispersion in oil, 4.81 g, 125 mmol) was added and stirred for 30 minutes. 2,3,4-Trifluoronitrobenzene (23.06 g, 130 mmol) was added and the reaction was allowed to warm to room temperature. Stirring was continued until TLC showed no remaining starting material. The reaction mixture was poured into a mixture of 10% HCl and ice. Ethyl acetate was added and the organic layer was separated, dried over $MgSO_4$, filtered and evaporated. The resulting oil was triturated with hexanes and Et_2O to afford a solid. The washings were collected, evaporated and triturated with hexanes. The materials were combined to afford the title compound (30 g, 74%) as a solid. ^{1}H NMR (400 MHz, $CDCl_3$): δ ppm 7.98 (ddd, 1 H), 7.43 (t, 1 H), 7.34 (ddd, 1 H), 7.21 (t, 1 H), 7.08 (dd, 1H).

Compound 5. 3-chloro-5-[(2-fluoro-3-methyl-6-nitrophenyl)oxy]benzonitrile. Sodium hydride (60% dispersion in oil, 2.16 g, 54.1 mmol) was added to anhydrous THF (50 mL) and cooled to 0 °C under

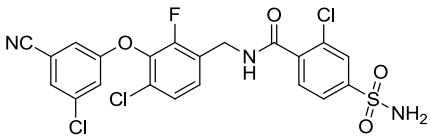
nitrogen. Di-*tert*-butyl malonate (4.65 g, 24.8 mmol) was added dropwise and the reaction mixture was stirred 15 min and allowed to warm to room temperature. The reaction was cooled to 0 °C and 3-chloro-5-[(2,3-difluoro-6-nitrophenyl)oxy]benzonitrile (7.0 g, 22.5 mmol) was added dropwise in a minimal amount of THF. The reaction was allowed to warm to room temperature and stirred for 3 hours then 10% aqueous citric acid (50 ml) and EtOAc (50 mL) were added. The layers were separated and the aqueous layer was extracted with EtOAc (2 x 50 mL). The organic extracts were combined, washed with brine, dried over Na_2SO_4, filtered and evaporated to afford bis(1,1-dimethylethyl) {3-[(3-chloro-5-cyanophenyl)oxy]-2-fluoro-4-nitrophenyl}propanedioate as an oil. ^{1}H NMR (400 MHz, DMSO-d_6): δ ppm 8.13 (dd, 1 H), 7.84 (d, 1 H), 7.65 (dd, 1 H), 7.59 - 7.61 (m, 1 H), 7.56 - 7.59 (m, 1 H), 5.10 (s, 1 H), 1.40 (s, 18 H). MS: *m/z* 507.2 (M+1).
The crude bis(1,1-dimethylethyl) {3-[(3-chloro-5-cyanophenyl)oxy]-2-fluoro-4-nitrophenyl}propanedioate was dissolved in CH_2Cl_2 (25 mL) and TFA (25 mL) and heated to reflux for 2h. The reaction mixture was cooled to room temperature and evaporated. Water (50 mL) and EtOAc (50 mL) were added, the layers were separated and the aqueous layer extracted with EtOAc (3 x 50 mL). The organic extracts were combined, dried over Na_2SO_4, filtered and evaporated to give {3-[(3-chloro-5-cyanophenyl)oxy]-2-fluoro-4-nitrophenyl}acetic acid as an oil. ^{1}H NMR (400 MHz, DMSO-*d*6): δ ppm 8.03 (d, 1 H), 7.81 (s, 1 H), 7.50 - 7.62 (m, 3 H), 3.84 (s, 2 H). MS: *m/z* 349.1 (M-1).
The crude {3-[(3-chloro-5-cyanophenyl)oxy]-2-fluoro-4-nitrophenyl}acetic acid was dissolved in CH_3CN (50 mL) and Cu_2O (0.65 g, 4.5 mmol) was added. A condenser was attached and the heterogeneous mixture was heated to reflux for 2 hours. The reaction mixture was cooled to room temperature, filtered through Celite and the solvent evaporated. Purification was accomplished by column chromatography (hexane/EtOAc) to afford the title compound (6.0 g, 87%), which solidified upon standing. ^{1}H NMR (400 MHz, DMSO-d_6): δ ppm 7.98 (dd, 1 H), 7.81 (d, 1 H), 7.60 (d, 2 H), 7.52 (t, 1 H), 2.37 (d, 3 H).

Compound 6. 3-{[3-(aminomethyl)-6-chloro-2-fluorophenyl]oxy}-5-chlorobenzonitrile. Sodium hydrosulfite (10.2 g, 58.7 mmol) dissolved in water (66 ml) was added dropwise to a vigorously stirred solution of 3-chloro-5-[(2-fluoro-3-methyl-6-nitrophenyl)oxy]benzonitrile (3.0 g, 9.78 mmol) dissolved in THF (33 mL). The reaction was stirred for 1 hour then EtOAc (100mL) was added and the layers were separated. The aqueous layer was extracted with EtOAc (2 x 50 mL). The organic extracts were combined, dried over Na_2SO_4, filtered and evaporated to afford the 3-[(6-amino-2-fluoro-3-methylphenyl)oxy]-5-chlorobenzonitrile (2.5 g, 92%) as a solid. ^{1}H NMR (400 MHz, DMSO-d_6): δ ppm 7.68 (d, 1H), 7.26 (dd, 1 H), 7.16 (t, 1 H), 6.85 (t, 1 H), 6.51 (dd, 1 H), 5.17 (d, 2 H), 2.06 (d, 3H) MS: *m/z* 277.2 (M+1).
To an oven dried flask was added $CuCl_2$ (2.5 g, 18.0 mmol). The flask was placed under high vacuum, flushed with nitrogen and acetonitrile (10 ml) was added. *t*-Butyl nitrite (2.7 mL, 22.5 mmol) was added dropwise. The stirred solution was placed in an oil bath at 50 °C under gentle stream of nitrogen and 3-[(6-amino-2-fluoro-3-methylphenyl)oxy]-5-chlorobenzonitrile dissolved in acetonitrile (15 mL) was added dropwise. The reaction was stirred for 0.5 hour, cooled to room temperature and poured into ice cold and aqueous HCl (0.5 N, 100 mL). EtOAc (100 mL) was added and the layers were separated. The aqueous layer was extracted with EtOAc (2 x 100 mL). The organic extracts were combined, dried over Na_2SO_4, filtered and evaporated. Purification was accomplished by silica gel column chromatography (hexane/EtOAc) to afford 3-chloro-5-[(6-chloro-2-fluoro-3-methylphenyl)oxy]benzonitrile (1.8 g, 67%) as a solid. ^{1}H NMR (400 MHz, DMSO-d_6): δ ppm 7.79 (s, 1H), 7.48 (s, 1 H), 7.44 (t, 1 H), 7.41 (d, 1 H), 7.31 (t, 1 H), 2.27 (d, 3 H).

3-Chloro-5-[(6-chloro-2-fluoro-3-methylphenyl)oxy]benzonitrile (1.8 g, 6.1 mmol) was dissolved in CCl_4 (200 mL). NBS (1.2 g, 6.7 mmol) and AIBN (0.63 g, 3.8 mmol) were added and the reaction mixture was placed in oil bath at 85 °C and stirred for 4 hours. The reaction was monitored by TLC and more AIBN

and NBS were added as necessary. The reaction was cooled, filtered and the solid washed with CCl_4. The solvent was evaporated. Purification was accomplished by silica gel column chromatography (hexane/EtOAc eluent) to afford 3-{[3-(bromomethyl)-6-chloro-2-fluorophenyl]oxy}-5-chlorobenzonitrile (1.8 g, 79%) as a solid. ^{1}H NMR(400 MHz, DMSO-d_6): δ ppm 7.81 (s, 1 H), 7.49 - 7.59 (m, 3 H), 7.47 (d, 1 H), 4.73 (s, 2 H). GC-MS, 372.9 (M).

3-{[3-(Bromomethyl)-6-chloro-2-fluorophenyl]oxy}-5-chlorobenzonitrile (1.0 g, 2.6 mmol) was dissolved in CH_2Cl_2 (5 mL) and added dropwise to ammonia in methanol (7N, 25 mL). The reaction mixture was stirred for 2 hours then the solvent was evaporated. The resulting oil was purified by silica gel column chromatography (CH_2Cl_2/MeOH) to afford 3-{[3-(aminomethyl)-6-chloro-2-fluorophenyl]oxy}-5-chlorobenzonitrile (0.7 g, 84%). ^{1}H NMR (400 MHz, DMSO-d_6): δ ppm 7.79 (s, 2 H), 7.45 - 7.56 (m, 4 H), 7.42 (t, 1 H), 3.75 (s, 2 H). MS: *m/z* 311.1 (M+1).



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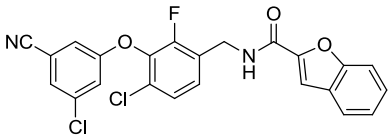
Compound 7. 4-(aminosulfonyl)-2-chloro-*N*-({4-chloro-3-[(3-chloro-5-cyanophenyl)oxy]-2-fluorophenyl}methyl)benzamide. 3-{[3-Aminomethyl)-6-chloro-2-fluorophenyl]oxy}-5-chlorobenzonitrile (0.030 g, 0.096 mmol), 4-(aminosulfonyl)-2-chlorobenzoic acid (0.034 g, 0.145 mmol) (prepared as described in WO2004054581), HATU (0.055 g, 0.145 mmol) and DIPEA (0.025 mL,0.145 mmol) were dissolved in DMF (2 mL) and stirred overnight. Purification was accomplished by Reverse-Phase HPLC (water/acetonitrile with 0.1%TFA). The desired fractions were neutralized with saturated $NaHCO_3$ and extracted with EtOAc (3 x 5 mL). The organic extracts were combined, dried over Na_2SO_4, filtered and evaporated to afford the title compound (0.025 g, 49%) as a white solid. ^{1}H NMR (400 MHz, DMSO-d_6): δ ppm 9.17 (t, 1 H), 7.86 (d, 1 H), 7.80 (d, 2 H), 7.64 (d, 1 H), 7.58 (br. s., 2 H), 7.48 - 7.55 (m, 2 H), 7.39 - 7.47 (m, 2 H), 4.50 (d, 2 H). MS: *m/z* 528.1 (M+1).



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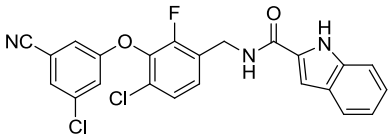
Compound 8. ***N*-({4-chloro-3-[(3-chloro-5-cyanophenyl)oxy]-2-fluorophenyl}methyl)-1-benzofuran-2-carboxamide (trifluoroacetate salt).** *N*-({4-Chloro-3-[(3-chloro-5-cyanophenyl)oxy]-2-fluorophenyl}methyl)-1-benzofuran-2-carboxamide trifluoroacetate was prepared in a similar manner as described herein from 3-{[3-aminomethyl)-6-chloro-2-fluorophenyl]oxy}-5-chlorobenzonitrile (0.025 g, 0.08 mmol), 1-benzofuran-2-carboxylic acid (0.020 g, 0.12 mmol), HATU (0.045 g, 0.12 mmol), DIPEA (0.020 mL, 0.12 mmol) and DMF (1 mL). Purification was accomplished by Reverse-Phase HPLC (water/acetonitrile with 0.1%TFA) to afford the title compound (0.024 g, 65%) as a white solid. ^{1}H NMR (400 MHz, DMSO-d_6): δ ppm 9.32 (t, 1 H), 7.82 (s, 1 H), 7.78 (d, 1 H), 7.66 (d, 1 H), 7.59 (s, 1 H), 7.44 - 7.55 (m, 4 H), 7.41 (t, 1 H), 7.34 (t, 1 H), 4.55 (d, 2 H). MS: *m/z* 455.0 (M+1).



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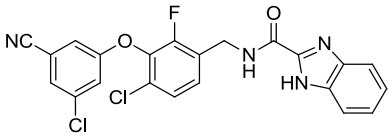
Compound 9. *N*-({4-chloro-3-[(3-chloro-5-cyanophenyl)oxy]-2-fluorophenyl}methyl)-1*H*-indole-2-carboxamide . *N*-({4-Chloro-3-[(3-chloro-5-cyanophenyl)oxy]-2-fluorophenyl}methyl)-1*H*-indole-2-carboxamide was prepared in a similar manner as described herein from 3-{[3-aminomethyl)-6-chloro-2-fluorophenyl]oxy}-5-chlorobenzonitrile (0.050 g, 0.16 mmol), indole-2-carboxylic acid (0.040 g, 0.24 mmol), HATU (0.090 g, 0.12 mmol), DIPEA(0.020 mL, 0.12 mmol) and DMF (2 mL). The crude material was purified by Reverse-Phase HPLC (water/acetonitrile with 0.1% TFA). The desired fractions were neutralized and extracted with EtOAc. The organic extracts were combined, dried over Na2SO4, filtered and evaporated to afford the title compound (0.052 g, 71%) as a white solid. ^{1}H NMR (400 MHz, DMSO-d_6): δ ppm 11.65 (s, 1 H), 9.10 (t, 1 H), 7.82 (s, 1 H), 7.62 (d, 1 H), 7.46 - 7.59 (m, 3 H), 7.34 - 7.46 (m, 2 H), 7.15 - 7.24 (m, 2 H), 7.04 (t, 1 H), 4.58 (d, 2 H). MS: *m/z* 454.1 (M+1).



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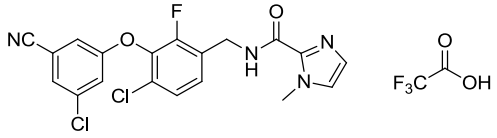
Compound 10. *N*-({4-chloro-3-[(3-chloro-5-cyanophenyl)oxy]-2-fluorophenyl}methyl)-1*H*-benzimidazole-2-carboxamide. *N*-({4-Chloro-3-[(3-chloro-5-cyanophenyl)oxy]-2-fluorophenyl}methyl)-1*H*-benzimidazole-2-carboxamide was prepared in a similar manner as described herein from 3-{[3-aminomethyl)-6-chloro-2-fluorophenyl]oxy}-5-chlorobenzonitrile (0.05 g, 0.16 mmol), 1*H*-benzimidazole-2-carboxylic acid (0.04 g, 0.24 mmol), HATU (0.09 g, 0.24 mmol), DIPEA (0.04 mL, 0.24 mmol) and DMF (2 mL). The crude material was purified by Reverse-Phase HPLC (water/acetonitrile with 0.1% TFA). The desired fractions were neutralized and extracted with EtOAc. The organic extracts were combined, dried over Na_2SO_4, filtered and evaporated to afford the title compound (0.035 g, 48 %) as a white solid. ^{1}H NMR (400 MHz, DMSO-d_6): δ ppm 9.57 (t, 1 H), 7.82 (t, 1 H), 7.57 - 7.72 (m, 2 H), 7.54 (d, 1 H), 7.47 - 7.52 (m, 2 H), 7.41 (t, 1 H), 7.30 (dd, 2 H), 4.58 (d, 2 H). MS: *m/z* 455.0 (M+1).



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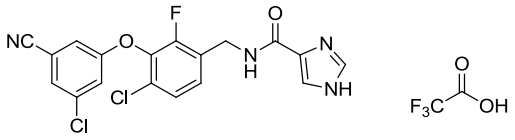
Compound 11. *N*-({4-chloro-3-[(3-chloro-5-cyanophenyl)oxy]-2-fluorophenyl}methyl)-1-methyl-1*H*-imidazole-5-carboxamide trifluoroacetate. *N*-({4-Chloro-3-[(3-chloro-5-cyanophenyl)oxy]-2-fluorophenyl}methyl)-1-methyl-1*H*-imidazole-5-carboxamide trifluoroacetate was prepared in a similar manner as described herein from 3-{[3-aminomethyl)-6-chloro-2-fluorophenyl]oxy}-5-chlorobenzonitrile (0.025 g, 0.08 mmol), 1-methyl-1*H*-imidazole-5-carboxylic acid (0.015 g, 0.12 mmol), HATU (0.045 g, 0.12 mmol), DIPEA (0.020 mL, 0.12 mmol) and DMF (1 mL). Purification was accomplished by Reverse-Phase HPLC (water/acetonitrile with 0.1%TFA). The desired fractions were lyophilized to afford the title compound (0.035 g, 83%) as a white solid. ^{1}H NMR (400 MHz, DMSO-d_6): δ ppm 9.13 (t, 1 H), 8.62 (br. s., 1 H), 7.95 (s, 1 H), 7.81 (s, 1 H), 7.43 - 7.52 (m, 3 H), 7.38 (t, 1 H), 4.48 (d, 2 H), 3.90 (s, 3 H). MS: *m/z* 419.0 (M+1).



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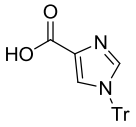
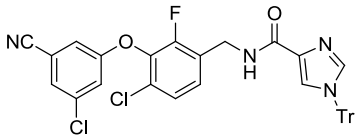
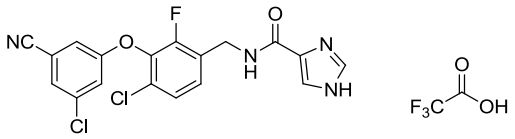
Compound 12. *N*-({4-chloro-3-[(3-chloro-5-cyanophenyl)oxy]-2-fluorophenyl}methyl)-1*H*-imidazole-4-carboxamide trifluoroacetate.



Step A: 1-(Triphenylmethyl)-1*H*-imidazole-4-carboxylic acid. 1-(Triphenylmethyl)-1*H*-imidazole-4-carboxylic acid was prepared as described in *J. Med. Chem*. **2001**, *44*, 1268. 1*H*-Imidazole-4-carboxylic acid (0.50 g, 4.5 mmol) and trityl chloride (1.35 g, 4.9 mmol) were added to a solution of DMF (30 mL) and pyridine (15 mL) and stirred overnight. Water and EtOAc were added. The layers were separated and the aqueous layer extracted with EtOAc (2 x 50 mL). The organic extracts were combined, washed with water and brine, dried over Na_2SO_4, filtered and evaporated. The oil was triturated with EtOAc to afford the title compound (1.5 g, 95%) as a white solid. ^{1}H NMR (400 MHz, DMSO-d_6): δ ppm 12.40 (br. s., 1 H), 7.42 (t, 9 H), 7.17 - 7.35 (m, 2 H), 7.10 (d, 6 H). MS: *m/z* 111 (M-243).



Step B: *N*-({4-chloro-3-[(3-chloro-5-cyanophenyl)oxy]-2-fluorophenyl}methyl)-1-(triphenylmethyl)-1*H*-imidazole-4-carboxamide. 3-{[3-Aminomethyl)-6-chloro-2-fluorophenyl]oxy}-5-chlorobenzonitrile (0.100 g, 0.30 mmol), 1-(triphenylmethyl)-1*H*-imidazole-4-carboxylic acid (0.140 g, 0.40 mmol), HATU (0.150 g, 0.40 mmol) and DIPEA (0.070 mL, 0.40 mmol) were dissolved in DMF (5 mL) and stirred overnight. The solvent was evaporated. Water and EtOAc were added. The layers were separated and the aqueous layer extracted with EtOAc. The organic extracts were combined, dried over Na_2SO_4, filtered and evaporated. Purification was accomplished by column chromatography (hexane/EtOAc) to afford the title compound (0.150 g, 72%) as a white solid. ^{1}H NMR (400 MHz, DMSO-d_6): δ ppm 8.70 (s, 1 H), 7.81 (t, 1 H), 7.55 (d, 1 H), 7.40 - 7.51 (m, 12 H), 7.30 - 7.36 (m, 2 H), 7.12 (dd, 6 H), 4.45 (d, 2 H). MS: *m/z* 649.2 (M+1).



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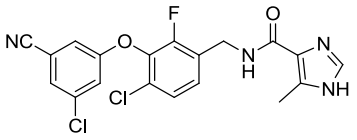
Step C. *N*-({4-chloro-3-[(3-chloro-5-cyanophenyl)oxy]-2-fluorophenyl}methyl)-1*H*-imidazole-4-carboxamide trifluoroacetate. *N*-({4-Chloro-3-[(3-chloro-5-cyanophenyl)oxy]-2-fluorophenyl}methyl)-1-(triphenylmethyl)-1*H*-imidazole-4-carboxamide (0.145 g, 0.22 mmol) was dissolved in CH_2Cl_2 (3 mL) and TFA (3 mL) added. The solution was stirred for 2.5 hours. The solvent was evaporated. Purification was accomplished by Reverse-Phase HPLC (water/acetonitrile with 0.1% TFA) to afford the title compound (0.070 g, 60%) as a white solid. ^{1}H NMR (400 MHz, DMSO-d_6): δ ppm 9.01 (br. s., 1 H), 8.53 (br. s., 1 H), 7.94 (s, 1 H), 7.83 (s, 1 H), 7.45 - 7.54 (m, 4 H), 7.38 (t, 1 H), 4.54 (d, 2 H). MS: *m/z* 405.0 (M+1).



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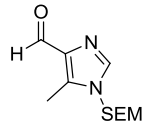
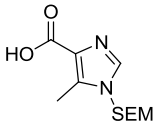
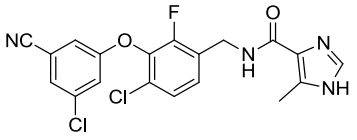
Compound 13. N-({4-chloro-3-[(3-chloro-5-cyanophenyl)oxy]-2-fluorophenyl}methyl)-4-methyl-1H-imidazole-5-carboxamide.



Step A: 5-methyl-1-({[2-(trimethylsilyl)ethyl]oxy}methyl)-1*H*-imidazole-4-carbaldehyde (+ isomer). A solution of 4-methyl-1*H*-imidazole-5-carbaldehyde (1.00 g, 9.1 mmol) in DMF (25 mL) was added dropwise to a 0 °C solution of NaH (0.36 g of a 60% dispersion in mineral oil, 9.1 mmol) in DMF (10 mL). The reaction mixture was stirred for 2 hours at room temperature, cooled to 0 °C and treated with a solution of SEMCl (1.6 mL, 9.1 mmol) in THF (10 mL). After stirring overnight at room temperature, the solution was quenched by the addition of water and extracted with CH_2Cl_2. The organic phase was dried (Na_2SO_4), filtered, and purified by silica gel chromatography (10-70% EtOAc/hexanes) to provide the title compound (2.11 g, 97%) as a clear oil and as a mixture of expected isomers. Major isomer: ^{1}H NMR (400 MHz, CHLOROFORM-*d*) δ ppm 9.89 (s, 1 H), 7.84 (s, 1 H), 5.67 (s, 2 H), 3.57 - 3.65 (m, 2 H), 2.57 (s, 3 H), 0.87 - 0.99 (m, 2 H), 0.00 (d, *J*=0.73 Hz, 9 H).



Step B: 5-methyl-1-({[2-(trimethylsilyl)ethyl]oxy}methyl)-1*H*-imidazole-4-carboxylic acid. The title compound was obtained from 5-methyl-1-({[2-(trimethylsilyl)ethyl]oxy}methyl)-1*H*-imidazole-4-carbaldehyde using a procedure and process similar to that described herein. The title compound, which was a single isomer, precipitated out of solution as a white solid (0.48 g, 45%) and was collected by filtration. ^{1}H NMR (400 MHz, DMSO-d_6) δ ppm 7.77 (s, 1 H), 5.33 (s, 2 H), 3.46 (t, *J*=7.90 Hz, 2 H), 2.46 (s, 3 H), 0.84 (t, *J*=7.90 Hz, 2 H), -0.05 (s, 9 H).



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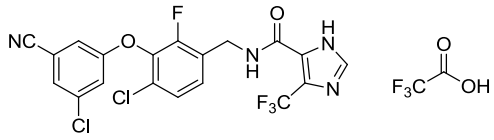
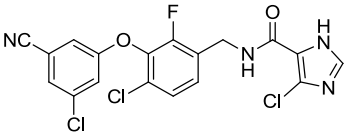
Step C: *N*-({4-chloro-3-[(3-chloro-5-cyanophenyl)oxy]-2-fluorophenyl}methyl)-4-methyl-1*H*-imidazole-5-carboxamide. The procedure and process are similar to that described herein except that 5-methyl-1-({[2-(trimethylsilyl)ethyl]oxy}methyl)-1*H*-imidazole-4-carboxylic acid (0.077 g, 0.30 mmol) and 3-{[3-(aminomethyl)-6-chloro-2-fluorophenyl]oxy}-5-chlorobenzonitrile (0.093 g, 0.30 mmol) were employed to provide the title compound (0.32 g, 25% overall) as a white solid after deprotection and purification by Reverse-Phase HPLC (water:acetonitrile with 0.1% NH_4OH. ^{1}H NMR (400 MHz, DMSO-d_6) δ ppm 12.29 (br. s., 1 H), 8.43 (t, *J*=5.84 Hz, 1 H), 7.82 (s, 1 H), 7.57 (s, 1 H), 7.44 - 7.54 (m, 3 H), 7.32 (t, *J*=7.83 Hz, 1 H), 4.45 (d, *J*=6.04 Hz, 2 H), 2.43 (s, 3 H). ES-LCMS: m/z 419.0, 421.0 (M+1).



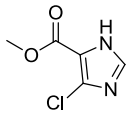
14

Compound 14. *N*-({4-chloro-3-[(3-chloro-5-cyanophenyl)oxy]-2-fluorophenyl}methyl)-1*H*-pyrrolo[2,3-*c*]pyridine-2-carboxamide trifluoroacetate. To a solution of ethyl 4-(trifluoromethyl)-1*H*-imidazole-5-carboxylate (0.030 g, 0.144 mmol) in THF/MeOH/H_2O (1:1:1, 1.5 mL) was added lithium hydroxide (0.034 g, 1.44 mmol) and the solution stirred at room temperature for 2 hours. A 10% solution of citric acid (1 mL) and EtOAc (5 mL) were added. The aqueous layer was separated and extracted with EtOAc (2 x 5 mL). The organic layers were combined, dried over sodium sulfate, filtered, concentrated and placed under high vacuum to afford the crude carboxylic acid. To the crude intermediate was added HATU (0.050 g, 0.144 mmol), DIPEA (0.025 mL, 0.144 mmol), 3-{[3-aminomethyl)-6-chloro-2-fluorophenyl]oxy}-5-chlorobenzonitrile (0.030 g, 0.144 mmol), and DMF (1 mL). The reaction mixture was stirred overnight. Purification was accomplished by RPHPLC (water:acetonitrile with 0.1% TFA). The desired fractions were neutralized and extracted with EtOAc. The combined organics were dried over Na_2SO_4, filtered and concentrated to afford *N*-({4-chloro-3-[(3-chloro-5-cyanophenyl)oxy]-2-fluorophenyl}methyl)-4-(trifluoromethyl)-1*H*-imidazole-5-carboxamide (0.009 g, 13%) as a white solid. ^{1}H NMR (400 MHz, DMSO-d_6): δ ppm 8.74 - 8.91 (m, 1 H) 7.86 - 8.02 (m, 1 H) 7.80 (d, 1 H) 7.48 (s, 2 H) 7.44 (s, 1 H) 7.30 - 7.42 (m, 1 H) 4.47 (d, 2 H). MS: *m/z* 473.2 (M+1).

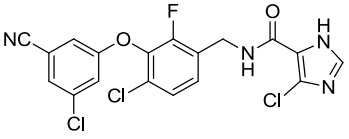


15

Compound 15. Example 131:4-chloro-*N*-({4-chloro-3-[(3-chloro-5-cyanophenyl)oxy]-2-fluorophenyl}methyl)-1*H*-imidazole-5-carboxamide.

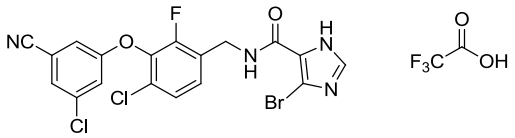


Step A: methyl 4-chloro-1H-imidazole-5-carboxylate. *N*-chlorosuccinimide (0.318 g, 2.4 mmol) was added to a stirred solution of methyl 1H-imidazole-4-carboxylate (0.300 g, 2.4 mmol) in CH_3CN (16 mL). The reaction mixture was stirred for 12 hours in the dark and then concentrated. The white residue was taken up in EtOAc, satd. aqueous Na_2S (10 mL) was added and the solution was stirred for 15 min. The organic layer was isolated, dried (Na_2SO_4), concentrated and the crude product was purified by column chromatography (20-70% EtOAc/hexanes) to afford the title compound (0.061 g, 16%) as a white solid. ^{1}H NMR (400 MHz, DMSO-d_6) δ ppm 11.05 (br. s., 1 H), 7.85 (s, 1 H), 3.81 (s, 3 H). ES-LCMS: *m/z* 160.9, 162.9 (M+1).



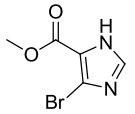
15

Step B: 4-chloro-*N*-({4-chloro-3-[(3-chloro-5-cyanophenyl)oxy]-2-fluorophenyl}methyl)-1*H*-imidazole-5-carboxamide. A solution of methyl 4-chloro-1*H*-imidazole-5-carboxylate (0.061 g, 0.4 mmol) was stirred in a solution of MeOH (2 mL), dioxane (2 mL) and 2N NaOH (1.9 mL, 3.8 mmol) at room temperature for 4 days. The reaction mixture was then acidified by addition of 1M HCl (20 mL) and extracted with EtOAc. The organic extracts were dried (Na_2SO_4), and the solvent was evaporated to provide 4-chloro-1H-imidazole-5-carboxylic acid (0.0523g, 94%) as a white solid which was used without further purification. 4-Chloro-*N*-({4-chloro-3-[(3-chloro-5-cyanophenyl)oxy]-2-fluorophenyl}methyl)-1*H*-imidazole-5-carboxamide was prepared in a similar manner as described herein from 3-{[3-aminomethyl)-6-chloro-2-fluorophenyl]oxy}-5-chlorobenzonitrile (0.054 g, 0.17 mmol), 4-chloro-1H-imidazole-5-carboxylic acid (0.026 g, 0.17 mmol), HATU (0.086 g, 0.23 mmol), DIPEA (0.039 mL, 0.23 mmol) and DMF (2 mL). Purification was accomplished by Reverse-Phase HPLC (water/acetonitrile with 0.1% TFA) to afford the title compound (0.008 g, 10%) as a white solid. ^{1}H NMR (400 MHz, DMSO-d_6): δ ppm 13.16 (br. s., 1 H), 8.24 (br. s., 1 H), 7.82 (s, 1 H), 7.78 (s, 1 H), 7.45 - 7.54 (m, 3 H), 7.35 - 7.42 (m, 1 H), 4.54 (s, 2 H). ES-LCMS: *m/z* 439.0, 441.0 (M+1).

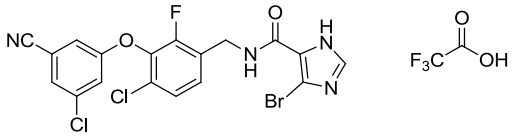


16

Compound 16. Example 246: 4-bromo-*N*-({4-chloro-3-[(3-chloro-5-cyanophenyl)oxy]phenyl}methyl)-1*H*-imidazole-5-carboxamide.



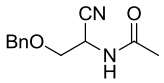
Step A: methyl 4-bromo-1*H*-imidazole-5-carboxylate. *N*-bromosuccinimide (0.900 g, 5.0 mmol) was added to a stirred solution of methyl 1*H*-imidazole-4-carboxylate (0.630 g, 5.0 mmol) in CH_3CN (50 mL). The reaction mixture was stirred for 12 hours in the dark and then concentrated in the presence of silica gel. The absorbed crude material was purified by column chromatography (20-100% EtOAc/hexanes) to afford the title compound (0.708 g, 70%) as a white solid. ^{1}H NMR (400 MHz, DMSO-d_6) δ ppm 11.00 (br. s., 1 H), 7.81 (s, 1 H), 3.81 (s, 3 H). ES-LCMS: *m/z* 204.9, 206.9 (M+1).



16

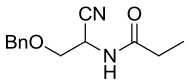
Step B: 4-bromo-*N*-({4-chloro-3-[(3-chloro-5-cyanophenyl)oxy]-2-fluorophenyl}methyl)-1*H*-imidazole-5-carboxamide trifluoroacetate. A solution of methyl 4-bromo-1*H*-imidazole-5-carboxylate (0.708 g, 3.5 mmol) was stirred in a solution of MeOH (20 mL) and 2N NaOH (20 mL, 40 mmol) at 40 °C for 4 hours, then additional 2N NaOH (20 mL, 40 mmol) was added and the reaction mixture kept at 40 °C for another hour. The reaction mixture was then acidified by addition of 1M HCl (100 mL) and extracted with EtOAc. The organic extracts were dried (Na_2SO_4), and the solvent was evaporated to provide crude

4-bromo-1*H*-imidazole-5-carboxylic acid (0.156g, 24%) as a white solid which was used without further purification.
4-Bromo-*N*-({4-chloro-3-[(3-chloro-5-cyanophenyl)oxy]-2-fluorophenyl}methyl)-1*H*-imidazole-5-carboxamide trifluoroacetate was prepared in a similar manner as described herein from 3-{[3-aminomethyl)-6-chloro-2-fluorophenyl]oxy}-5-chlorobenzonitrile (0.062 g, 0.20 mmol), 4-bromo-1*H*-imidazole-5-carboxylic acid (0.038 g, 0.20 mmol), HATU (0.091 g, 0.24 mmol), DIPEA (0.042 mL, 0.24 mmol) and DMF (2 mL). Purification was accomplished by Reverse-Phase HPLC (water/acetonitrile with 0.1% TFA) to afford the title compound (0.010 g, 8%) as a white solid. ^{1}H NMR (400 MHz, DMSO-d_6) δ ppm 8.38 (br. s., 1 H), 7.82 (s, 2 H), 7.44 - 7.54 (m, 3 H), 7.38 (t, 1 H), 4.51 (d, 2 H). ES-LCMS: *m/z* 482.9, 484.9, 486.9 (M+1).



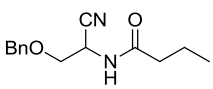
18

Compound 18. N-(2-(benzyloxy)-1-cyanoethyl)acetamide. Benzyloxyacetaldehyde (1.5 g, 10.0 mmol) was added dropwise to a stirred solution of sodium cyanide (0.61 g, 12.4 mmol) and ammonium chloride (0.79 g, 14.8 mmol) in 25% ammonium hydroxide (5 mL). The solution was stirred at room temperature for 48 hours, then extracted with CH_2Cl_2 (20 mL). The organic extract was washed with brine, dried (Na_2SO_4) and filtered. The filtrate was cooled to 0 °C, and pyridine (1.3 mL, 16.1 mmol) was added followed by acetyl chloride (1.1 mL, 15.5 mmol). The reaction mixture was stirred for 2 hours at 0 °C. Water was added (10 mL) and the organic phase was separated and washed with 1N HCl (20 mL), dried (Na_2SO_4), filtered and concentrated. The residue was purified by silica gel flash column chromatography (5-50% EtOAc : hexanes) to give the title compound (1.60 g, 65%) as an off-white solid. ^{1}H NMR (400 MHz, $CDCl_3$) δ ppm 7.33 - 7.44 (m, 5 H), 6.14 (br. s., 1 H), 5.05 - 5.12 (m, 1 H), 4.65 (ABq, 2 H), 3.69 (ABq, 2 H), 2.04 (s, 3 H).



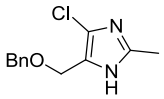
19

Compound 19. *N*-{1-cyano-2-[(phenylmethyl)oxy]ethyl}propanamide. Benzyloxyacetaldehyde (3.0 g, 20.0 mmol) was added dropwise to a stirred solution of sodium cyanide (1.22 g, 24.9 mmol) and ammonium chloride (1.58 g, 29.5 mmol) in 25% ammonium hydroxide (9.6 mL). The solution was stirred at room temperature for 48 hours, then extracted with CH_2Cl_2 (40 mL). The organic extract was washed with brine, dried (Na_2SO_4) and filtered. Propionic acid (1.5 g, 20.0 mmol), EDC (5.7 g, 30.0 mmol) and DMAP (610 mg, 5 mmol) were added to the filtrate and the solution was stirred for 12 hours at room temperature. Water was added (20 mL) and the organic phase was separated and washed with 1N HCl (20 mL), dried (Na_2SO_4), filtered and concentrated. The residue was purified by silica gel flash column chromatography (0-50% EtOAc : hexanes) to give the title compound (3.62 g, 78%) as a yellow oil. ^{1}H NMR (400 MHz, $CDCl_3$) δ ppm 7.32 - 7.43 (m, 5 H), 6.09 (br. s., 1 H), 5.08 - 5.13 (m, 1 H), 4.59 - 4.69 (ABq, 2 H), 3.66 (ABq, 2 H), 2.25 (qd, 2 H), 1.17 (t, 3 H). ES-LCMS: *m/z* 233.0 (M+H).



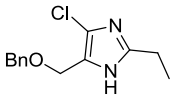
20

Compound 20. *N*-{1-cyano-2-[(phenylmethyl)oxy]ethyl}butanamide. Benzyloxyacetaldehyde (1.5 g, 10.0 mmol) was added dropwise to a stirred solution of sodium cyanide (0.61 g, 12.4 mmol) and ammonium chloride (0.79 g, 14.8 mmol) in 25% ammonium hydroxide (5 mL). The solution was stirred at RT for 48 hours, then extracted with CH_2Cl_2 (20 mL). The organic extract was washed with brine, dried (Na_2SO_4) and filtered. The filtrate was cooled to 0 °C, and pyridine (1.3 mL, 16.1 mmol) was added followed by butyryl chloride (1.56 mL, 15.0 mmol). The reaction mixture was stirred for 2 hours at 0 °C. Water was added (10 mL) and the organic phase was separated, dried (Na_2SO_4), filtered and concentrated. The residue was purified by silica gel flash column chromatography (5-35% EtOAc : hexanes) to give the title compound (1.99 g, 81%) as a light yellow oil. ^{1}H NMR (400 MHz, $CDCl_3$) δ ppm 7.32 - 7.43 (m, 5 H), 6.09 (d, 1 H), 5.08 - 5.14 (m, 1 H), 4.64 (ABq, 2 H), 3.71 (ABq, 2 H), 2.20 (t, 2 H), 1.62 - 1.73 (m, 2 H), 0.96 (t, 3 H). ES-LCMS: *m/z* 247.2 (M+1).



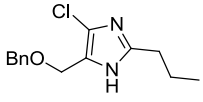
22

Compound 22. 4-chloro-2-methyl-5-{[(phenylmethyl)oxy]methyl}-1*H*-imidazole. A solution of *N*-{1-cyano-2-[(phenylmethyl)oxy]ethyl}acetamide (compound 18) (1.64 g, 7.5 mmol), triphenylphosphine (4.9 g, 18.8 mmol), and carbon tetrachloride (1.8 mL, 18.8 mmol) in acetonitrile (75 mL) was heated at 45 °C for 6 hours. The reaction was concentrated and the residue was stirred in CH_2Cl_2 (80 mL) and 0.5 N NaOH (70 mL) for 15 min. The organic layer was isolated, dried (Na_2SO_4), filtered, concentrated and the residue was purified by silica gel flash column chromatography (5-70% EtOAc : hexanes) to give the title compound as a yellow solid, in assumed quantitative yield due to inseparable triphenylphosphine oxide. ^{1}H NMR (400 MHz, $CDCl_3$) δ ppm 7.28 - 7.37 (m, 5 H), 4.50 (s, 2 H), 4.48 (s, 2 H), 2.37 (s, 3 H).



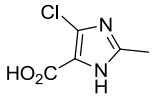
23

Compound 23. 4-chloro-2-ethyl-5-{[(phenylmethyl)oxy]methyl}-1*H*-imidazole. A solution of *N*-{1-cyano-2-[(phenylmethyl)oxy]ethyl}propanamide (compound 19) (3.62 g, 15.6 mmol), triphenylphosphine (10.2 g, 39.0 mmol), and carbon tetrachloride (3.8 mL, 39.0 mmol) in acetonitrile (150 mL) was heated at 45 °C for 4.5 hours. The reaction was concentrated and the residue was stirred in CH_2Cl_2 (170 mL) and 0.5 N NaOH (150 mL) for 15 min. The organic layer was isolated, dried (Na_2SO_4), filtered, concentrated and the residue was purified by silica gel flash column chromatography (5-50% EtOAc : hexanes) to give the title compound (2.51 g, 64%) as a light yellow solid. ^{1}H NMR (400 MHz, $CDCl_3$) δ ppm 7.29 - 7.38 (m, 5 H), 4.52 (s, 2 H), 4.50 (s, 2 H), 2.65 (q, 2 H), 1.26 (t, 3 H). ES-LCMS: *m/z* 250.6, 252.9 (M+H).



24

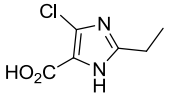
Compound 24. 4-chloro-5-{[(phenylmethyl)oxy]methyl}-2-propyl-1*H*-imidazole. A solution of *N*-{1-cyano-2-[(phenylmethyl)oxy]ethyl}butanamide (compound 20) (1.60 g, 6.5 mmol), triphenylphosphine (4.3 g, 16.3 mmol), and carbon tetrachloride (1.6 mL, 16.3 mmol) in acetonitrile (65 mL) was heated at 42 °C for 12 hours. The reaction was then concentrated and the residue was stirred in CH_2Cl_2 (70 mL) and 0.5 N NaOH (60 mL) for 15 min. The organic layer was isolated, dried (Na_2SO_4), filtered, concentrated and the residue was purified by silica gel flash column chromatography (5-40% EtOAc : hexanes) to give the title compound as a light yellow solid, in assumed quantitative yield due to inseparable triphenylphosphine oxide. ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.30 - 7.40 (m, 5 H), 4.52 (s, 4 H), 2.58 - 2.68 (m, 2 H), 1.68 - 1.82 (m, 2 H), 0.97 (t, 3 H).



25

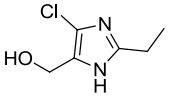
Compound 25. 4-chloro-2-methyl-1*H*-imidazole-5-carboxylic acid. A solution of 4-chloro-2-methyl-5-{[(phenylmethyl)oxy]methyl}-1*H*-imidazole (**22**) (1.78 g, 7.5 mmol) and methanesulfonic acid (18.5 mL, 285 mmol) in chloroform (39 mL) was stirred at room temperature for 1 hour. The reaction mixture was poured into ice (~70 g) and the solution was neutralized by addition of 5N NaOH until the pH was 10. The solution was extracted with methyl *tert*-butyl ether (2 x 60 mL) and then with *n*-butanol (3 x 40 mL). The combined *n*-butanol extracts were washed with brine, concentrated, and dried *in vacuo* to provide crude (4-chloro-2-methyl-1*H*-imidazol-5-yl)methanol as a brown solid. A solution of this solid and manganese dioxide (2.0 g, 23.0 mmol) in CH_2Cl_2 (12 mL) and 1,4-dioxane (6 mL) was heated under reflux for 6 hours. The reaction mixture was cooled to room temperature, and filtered through Celite, which was washed thoroughly with CH_2Cl_2. The filtrate was concentrated and dried to provide crude 4-chloro-2-methyl-1*H*-imidazole-5-carbaldehyde as a yellow solid (0.308 g, 28%), which was used without further purification.

A solution of sodium chlorite (1.4 g, 15.3 mmol) and sodium dihydrogen phosphate monohydrate (1.2 g, 8.7 mmol) in water (3.3 mL) was added to a stirred solution of 4-chloro-2-methyl-1*H*-imidazole-5-carbaldehyde (0.308 g, 2.1 mmol), 2-methyl-2-butene (9.3 mL of a 2M solution in THF, 18.6 mmol), and *tert*-butanol (1.2 mL) in THF (4.7 mL). The reaction mixture was stirred at room temperature for 6 hours. The aqueous phase was separated and extracted with EtOAc (4 x 10 mL). The combined extracts were dried (Na_2SO_4), filtered, concentrated and the residue was triturated with diethyl ether to provide the title compound (0.168 g, 50%) as white solid. ^{1}H NMR (400 MHz, DMSO-d_6) δ ppm 12.98 (br. s., 1 H), 12.88, (br. s., 1 H), 2.26 (s, 3 H). ES-LCMS: *m/z* 160.9 (M+1).

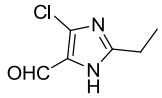


26

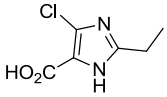
Compound 26: : 4-chloro-2-ethyl-1*H*-imidazole-5-carboxylic acid



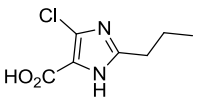
Step A: (4-chloro-2-ethyl-1*H*-imidazol-5-yl)methanol. A solution of 4-chloro-2-ethyl-5-{[(phenylmethyl)oxy]methyl}-1*H*-imidazole (**23**) (2.51 g, 10.0 mmol) and methanesulfonic acid (25 mL, 385 mmol) in chloroform (56 mL) was stirred at room temperature for 1 hour. The reaction mixture was poured into ice (~100 g) and the solution was neutralized by addition of 5N NaOH until the pH was 10. The solution was extracted with methyl *tert*-butyl ether (2 x 100 mL) and then with *n*-butanol (3 x 100 mL). The combined *n*-butanol extracts were concentrated, azeotroped with toluene, and dried *in vacuo* to provide 4-chloro-2-ethyl-1*H*-imidazol-5-yl)methanol in quantitative yield as an off-white solid. ^{1}H NMR (400 MHz, DMSO-d_6) δ ppm 12.10 (br. s., 1 H), 5.10 (br. s., 1 H), 4.32 (s, 2 H), 2.54 (q, 2 H), 1.15 (t, 3 H).



Step B: 4-chloro-2-ethyl-1*H*-imidazole-5-carbaldehyde. A solution of (4-chloro-2-ethyl-1*H*-imidazol-5-yl)methanol (1.60 g, 10.0 mmol) and manganese dioxide (5.5 g, 63.3 mmol) in CH_2Cl_2 (30 mL) and 1,4-dioxane (15 mL) was heated under reflux for 6.5 hours. The reaction mixture was cooled to room temperature, then filtered through Celite, which was washed thoroughly with CH_2Cl_2. The filtrate was concentrated and dried to provide the title compound (0.763 mg, 48%) as a pale yellow solid. ^{1}H NMR (400 MHz, $CDCl_3$) δ ppm 9.66 (s, 1 H), 2.84 (q, 2 H), 1.38 (t, 3 H).

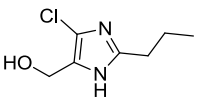


Step C: 4-chloro-2-ethyl-1*H*-imidazole-5-carboxylic acid. A solution of sodium chlorite (4.41 g, 48.8 mmol) and sodium dihydrogen phosphate monohydrate (3.9 g, 28.3 mmol) in water (11 mL) was added to a stirred solution of 4-chloro-2-ethyl-1*H*-imidazole-5-carbaldehyde (0.763 g, 4.81 mmol), 2-methyl-2-butene (30 mL of a 2M solution in THF, 60 mmol), and *tert*-butanol (3.7 mL) in THF (15 mL). The reaction mixture was stirred at room temperature for 6 hours. The aqueous phase was separated and extracted with EtOAc (4 x 40 mL). The combined extracts were dried (Na_2SO_4), filtered, concentrated and the residue was triturated with diethyl ether to provide the title compound (0.715 g, 85 %) as a white solid. ^{1}H NMR (400 MHz, DMSO-d_6) δ ppm 12.86 (br. s., 2 H), 2.59 (q, 2 H), 1.17 (t, 3 H). ES-LCMS: *m/z* 174.9, 176.9 (M+H).



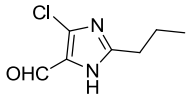
27

Compound 27: 4-chloro-2-propyl-1*H*-imidazole-5-carboxylic acid

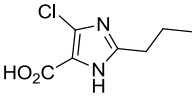


Step A: (4-chloro-2-propyl-1*H*-imidazol-5-yl)methanol. A solution of 4-chloro-5-{[(phenylmethyl)oxy]methyl}-2-propyl-1*H*-imidazole (1.72 g, 6.5 mmol) and methanesulfonic acid (16 mL, 247 mmol) in chloroform (36 mL) was stirred at room temperature for 1 hour. The reaction mixture

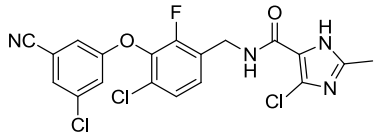
was poured into ice (~70 g) and the solution was neutralized by addition of 5N NaOH until the pH was 10. The solution was extracted with methyl *tert*-butyl ether (2 x 60 mL) and then with *n*-butanol (3 x 40 mL). The combined *n*-butanol extracts were concentrated, washed with brine, and dried *in vacuo* to provide the title compound (0.927 g, 82%) as a light yellow solid. ^{1}H NMR (400 MHz, DMSO-d_6) δ ppm 12.09 (br. s., 1 H), 5.10 (br. s., 1 H), 4.31 (s, 2 H), 2.45 - 2.51 (m, 2 H), 1.60 (sex, 2 H), 0.87 (t, 3 H).



Step B: 4-chloro-2-propyl-1*H*-imidazole-5-carbaldehyde. A solution of (4-chloro-2-propyl-1*H*-imidazol-5-yl)methanol (0.927 g, 5.3 mmol) and manganese dioxide (2 g, 23.0 mmol) in CH_2Cl_2 (12 mL) and 1,4-dioxane (6 mL) was heated under reflux for 6 hours. The reaction mixture was cooled to room temperature, then filtered through celite, washing thoroughly with CH_2Cl_2. The filtrate was concentrated and dried to provide the title compound (0.541 g, 59%) as a pale yellow solid. ^{1}H NMR (400 MHz, $CDCl_3$) δ ppm 11.61 (br. s., 1 H), 9.63 (s, 1 H), 2.80 (t, 2 H), 1.83 (sex, 2 H), 0.99 (t, 3 H). ES-LCMS: *m/z* 173.0 (M+1).

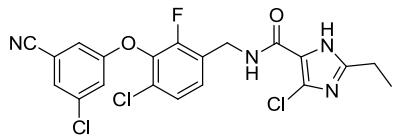


Step C: 4-chloro-2-propyl-1*H*-imidazole-5-carboxylic acid. A solution of sodium chlorite (2.9 g, 32.1 mmol) and sodium dihydrogen phosphate monohydrate (2.5 g, 18.1 mmol) in water (7.2 mL) was added to a stirred solution of 4-chloro-2-propyl-1*H*-imidazole-5-carbaldehyde (0.541 g, 3.1 mmol), 2-methyl-2-butene (19.3 mL of a 2M solution in THF, 38.6 mmol), and *tert*-butanol (2.3 mL) in THF (10 mL). The reaction mixture was stirred at room temperature for 6 hours. The aqueous phase was separated and extracted with EtOAc (4 x 40 mL). The combined extracts were dried (Na_2SO_4), filtered, concentrated and the residue was triturated with diethyl ether to provide the title compound (0.587 g, quant.) as a white solid. ^{1}H NMR (400 MHz, DMSO-d_6) δ ppm 12.99 (br. s., 1 H), 12.88, (br. s., 1 H), 2.55 (t, 2 H), 1.63 (sex, 2 H), 0.86 (t, 3 H). ES-LCMS: *m/z* 189.0 (M+H).



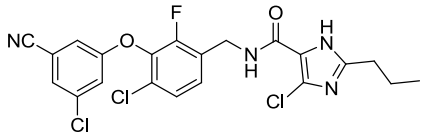
28

Compound 28. 4-chloro-*N*-({4-chloro-3-[(3-chloro-5-cyanophenyl)oxy]-2-fluorophenyl}methyl)-2-methyl-1*H*-imidazole-5-carboxamide. 4-chloro-*N*-({4-chloro-3-[(3-chloro-5-cyanophenyl)oxy]-2-fluorophenyl}methyl)-2-methyl-1*H*-imidazole-5-carboxamide was prepared in a similar manner as described herein from 3-chloro-5-({6-chloro-2-fluoro-3-[(methylamino)methyl]phenyl}oxy)benzonitrile (compound 25) (0.062 g, 0.20 mmol), 4-chloro-2-methyl-1*H*-imidazole-5-carboxylic acid (0.032 g, 0.20 mmol), HATU (0.091 g, 0.24 mmol), DIPEA (0.042 mL, 0.24 mmol) and DMF (2 mL). Purification was accomplished by Reverse-Phase HPLC (water/acetonitrile with 0.1% TFA) to afford the title compound (0.051 g, 56%) as a white solid. ^{1}H NMR (400 MHz, DMSO-d_6) δ ppm 12.77 (br. s., 1 H), 8.08 (br. s., 1 H), 7.80 - 7.84 (m, 1 H), 7.44 - 7.53 (m, 3 H), 7.36 (t, 1 H), 4.49 - 4.56 (m, 2 H), 2.26 (s, 3 H). ES-LCMS: *m/z* 453.0, 454.0 (M+1).



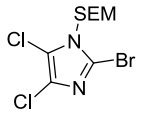
29

Compound 29. 4-chloro-*N*-({4-chloro-3-[(3-chloro-5-cyanophenyl)oxy]-2-fluorophenyl}methyl)-2-ethyl-1*H*-imidazole-5-carboxamide. 4-chloro-*N*-({4-chloro-3-[(3-chloro-5-cyanophenyl)oxy]-2-fluorophenyl}methyl)-2-ethyl-1*H*-imidazole-5-carboxamide was prepared in a similar manner as described herein from 3-chloro-5-({6-chloro-2-fluoro-3-[(methylamino)methyl]phenyl}oxy)benzonitrile (0.713 g, 2.29 mmol), 4-chloro-2-ethyl-1*H*-imidazole-5-carboxylic acid (compound 26) (0.400 g, 2.29 mmol), HATU (1.133 g, 2.98 mmol), DIPEA (0.520 mL, 2.98 mmol) and DMF (12 mL). Purification was accomplished by silica gel chromatography (0-5% MeOH (2M NH_3) in CH_2Cl_2) to afford the title compound (0.622 g, 58%) as a white solid. ^{1}H NMR (400 MHz, DMSO-d_6) δ 12.74 (s, 1 H), 8.09 (br. s., 1 H), 7.81 - 7.84 (m, 1 H), 7.48 - 7.53 (m, 2 H), 7.45 - 7.47 (m, 1 H), 7.33 - 7.40 (m, 1 H), 4.54 (d, 2 H), 2.60 (q, 2 H), 1.18 (t, 3 H). ES-LCMS: *m/z* 467.0, 469.0 (M+H).



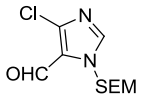
30

Compound 30. 4-chloro-*N*-({4-chloro-3-[(3-chloro-5-cyanophenyl)oxy]-2-fluorophenyl}methyl)-2-propyl-1*H*-imidazole-5-carboxamide. 4-chloro-*N*-({4-chloro-3-[(3-chloro-5-cyanophenyl)oxy]-2-fluorophenyl}methyl)-2-propyl-1*H*-imidazole-5-carboxamide was prepared in a similar manner as described herein from 3-chloro-5-({6-chloro-2-fluoro-3-[(methylamino)methyl]phenyl}oxy)benzonitrile (0.062 g, 0.20 mmol), 4-chloro-2-propyl-1*H*-imidazole-5-carboxylic acid (compound 27) (0.038 g, 0.20 mmol), HATU (0.091 g, 0.24 mmol), DIPEA (0.042 mL, 0.24 mmol) and DMF (2 mL). Purification was accomplished by Reverse-Phase HPLC (water/acetonitrile with 0.1% TFA) to afford the title compound (0.062 g, 64%) as a white solid. ^{1}H NMR (400 MHz, DMSO-d_6) δ ppm 12.73 (br. s., 1 H), 8.12 (br. s., 1 H), 7.81 - 7.84 (m, 1 H), 7.45 - 7.52 (m, 3 H), 7.37 (t, 1 H), 4.53 (d, 2 H), 2.55 (t, 2 H), 1.64 (sex, 2 H), 0.87 (t, 3 H). ES-LCMS: *m/z* 481.0, 483.0 (M+1).



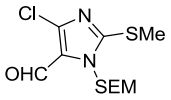
31

Compound 31. 2-bromo-4,5-dichloro-1-({[2-(trimethylsilyl)ethyl]oxy}methyl)-1H-imidazole. A solution of 2-bromo-4,5-dichloro-1H-imidazole (2.16 g, 10 mmol) in THF (25 mL) was added dropwise to a 0 ^{o}C suspension of NaH (440 mg of a 60 % suspension in mineral oil, 11 mmol) in THF (20 mL). The mixture was stirred at room temperature for 2 hours, cooled to 0 °C and {2-[(chloromethyl)oxy]ethyl}(trimethyl)silane (1.9 mL, 11 mmol) was added dropwise. The mixture was stirred at room temperature overnight. Saturated $NaHCO_3$ was added and the aqueous layer was extracted with CH_2Cl_2. The organic layer was dried (Na_2SO_4), filtered, concentrated and purified by silica gel chromatography (0-50% CH_2Cl_2/hexanes) to give the title compound (3.26 g, 95%) as a white solid. ^{1}H NMR (400 MHz, CHLOROFORM-*d*) δ ppm 5.26 (2 H, s) 3.55 - 3.61 (2 H, m) 0.88 - 0.94 (2 H, m) -0.02 (9 H, s). LCMS: m/z 346 (M+1).



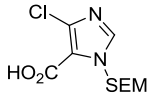
32

Compound 32. 5-chloro-1-((2-(trimethylsilyl)ethoxy)methyl)-1H-imidazole-4-carbaldehyde. nBuLi (3.74 ml of 2.4 M solution in hexanes, 8.99 mmol) was added dropwise to a -78 °C solution of bromoimidazole (3.11 g, 9.0 mmol) in tetrahydrofuran (THF) (18 ml). After 20 mins, TMS-Cl (1.14 ml, 9.0 mmol) was added dropwise. The reaction mixture was stirred for 15 mins, then removed from bath and allowed to warm to room temperature and stirred for 1 hour. The solution was then cooled to -78 °C, and nBuLi (4.12 ml of 2.4 M solution in hexanes, 9.9 mmol) was added dropwise. After 20 mins, DMF (3.48 ml, 44.9 mmol) was added. The reaction mixture was removed from the cooling bath and allowed to slowly warm to room temperature and quenched by the addition of saturated NH_4Cl. The organic layer isolated, dried (Na_2SO_4), filtered, and evaporated. The residue was taken up in EtOAc and 1N HCl and stirred for 10 mins. The organic layer was isolated and dried (Na_2SO_4), filtered, evaporated and purified by silica gel chromatography (0-30% EtOAc/hexanes) to afford the title compound (1.70 g, 73%) as a light yellow oil. ^{1}H NMR (400 MHz, CHLOROFORM-*d*) δ ppm 9.86 (s, 1 H), 7.75 (s, 1 H), 5.67 (s, 2 H), 3.57 - 3.67 (m, 2 H), 0.91 - 1.00 (m, 2 H), 0.00 (s, 9 H).



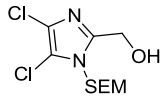
33

Compound 33. 4-chloro-2-(methylthio)-1-({[2-(trimethylsilyl)ethyl]oxy}methyl)-1H-imidazole-5-carbaldehyde. nBuLi (22.61 ml of 2.4 M solution in hexanes, 54.3 mmol) was added dropwise to a solution of 2-bromo-4,5-dichloro-1-({[2-(trimethylsilyl)ethyl]oxy}methyl)-1H-imidazole (18.78 g, 54.3 mmol) in tetrahydrofuran (THF) (200 ml) at -78 °C and the reaction mixture was stirred for 20 min. Dimethyl disulfide (4.82 ml, 54.3 mmol) was added dropwise slowly and stirring was continued for another 30 min. Additional nBuLi (22.61 ml of 2.4 M solution in hexanes, 54.3 mmol) was added to the solution and after another 20 min, DMF (8.40 ml, 109 mmol) was added slowly. The reaction mixture was stirred for 30 min, allowed to warm to room temperature, stirred overnight, and evaporated to half the volume. Saturated $NaHCO_3$ (50 mL) and EtOAc (50 mL) were added and the aqueous layer was extracted with EtOAc. The organic phase was dried (Na_2SO_4), filtered, and purified by silica gel chromatography (0-30% EtOAc/hex) to afford the title compound (12.59 g, 76 %) as a clear oil. ^{1}H NMR (400 MHz, CHLOROFORM-*d*) δ ppm 9.69 (s, 1 H), 5.66 (s, 2 H), 3.52 - 3.65 (m, 2 H), 2.71 (s, 3 H), 0.88 - 1.01 (m, 2 H), -0.01 (s, 9 H).



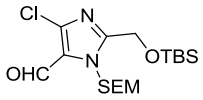
34

Compound 34: 4-chloro-1-({[2-(trimethylsilyl)ethyl]oxy}methyl)-1H-imidazole-5-carboxylic acid. The title compound (0.43 g, 77%) was obtained as a clear oil from 2-bromo-4,5-dichloro-1-({[2-(trimethylsilyl)ethyl]oxy}methyl)-1H-imidazole (0.69 g, 2.0 mmol) using a procedure and process similar to that described herein. ^{1}H NMR (400 MHz, DMSO-d_6) δ ppm 13.43 (br. s., 1 H), 8.10 (s, 1 H), 5.60 (s, 2 H), 3.48 (t, J=7.83 Hz, 2 H), 0.82 (t, J=7.83 Hz, 2 H), -0.06 (s, 9 H).



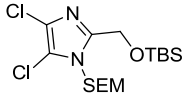
35

Compound 35. [4,5-Dichloro-1-({[2-(trimethylsilyl)ethyl]oxy}methyl)-1*H*-imidazol-2-yl]methanol. nBuLi (2.0 mL, 3.2 mmol, 1.6 M in hexane) was added dropwise to a solution of 2-bromo-4,5-dichloro-1-({[2-(trimethylsilyl)ethyl]oxy}methyl)-1*H*-imidazole (1.1 g, 3.2 mmol) in THF (50 mL) at -78 °C under N_2**.** The mixture was stirred for 30 min and paraformaldehyde (960 mg, 32 mmol) was then added. The mixture was allowed warm up to room temperature and stirred for 6 hours and then quenched by aqueous NH_4Cl. Extracted with EtOAc and the organic layer was dried over sodium sulfate and concentrated. The residue was purified by column chromatography (0-30% EtOAc in hexane) to give the title compound (614 mg, 65 %) as a colorless oil. ^{1}H NMR (400 MHz, CHLOROFORM-*d*) δ ppm 5.37 (2 H, s) 4.70 (2 H, d, *J*=1.2 Hz) 3.58 (2 H, dd, *J*=8.9, 7.7 Hz) 0.89 - 0.96 (2 H, m) 0.00 (9 H, s). LCMS: *m/z* 297 (M+1).

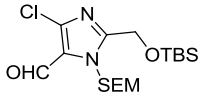


36

Compound 36. 2-(((tert-butyldimethylsilyl)oxy)methyl)-4-chloro-1-((2-(trimethylsilyl)ethoxy)methyl)-1H-imidazole-5-carbaldehyde.

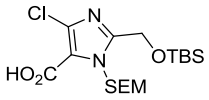


Step A. 2-(((tert-butyldimethylsilyl)oxy)methyl)-4,5-dichloro-1-((2-(trimethylsilyl)ethoxy)methyl)-1H-imidazole. Imidazole (282 mg, 4.14 mmol) was added to a solution of [4,5-dichloro-1-({[2-(trimethylsilyl)ethyl]oxy}methyl)-1*H*-imidazol-2-yl]methanol (**35**) (614 mg, 2.07 mmol) and chloro(1,1-dimethylethyl)dimethylsilane (345 mg, 2.28 mmol) in DCM (8 mL) at room temperature under N_2**.** The mixture was stirred overnight. Aqueous NH_4Cl was then added. After extracted with DCM, the combined organic layers were dried over sodium sulfate and concentrated. The residue was purified by column chromatography (0-10% EtOAc in hexane) to give the title compound (845 mg, 99 %) as a colorless oil. ^{1}H NMR (400 MHz, CHLOROFORM-*d*) δ ppm 5.42 (2 H, s) 4.74 (2 H, s) 3.53 - 3.59 (2 H, m) 0.87 - 0.95 (11 H, m) 0.09 (6 H, s) 0.00 (9 H, s). LCMS: *m/z* 411 (M+1).



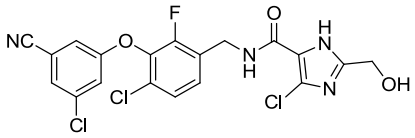
36

Step B. 2-(((tert-butyldimethylsilyl)oxy)methyl)-4-chloro-1-((2-(trimethylsilyl)ethoxy)methyl)-1H-imidazole-5-carbaldehyde. nBuLi (1.3 mL, 2.05 mmol, 1.57 M in hexane) was added dropwise to a solution of 4,5-dichloro-2-({[(1,1-dimethylethyl)(dimethyl)silyl]oxy}methyl)-1-({[2-(trimethylsilyl)ethyl]oxy}methyl)-1*H*-imidazole (842 mg, 2.05 mmol) in THF (30 mL) at -78 °C under N_2**.** The mixture was stirred for 30 min and DMF (1.0 mL) was then added dropwise. The mixture was stirred for 20 min and then allowed warm up to room temperature and stirred for 20 min. The reaction was quenched by aqueous NH_4Cl and extracted with EtOAc and the organic layer was dried over sodium sulfate and concentrated. The residue was purified by column chromatography (0-10% EtOAc in hexane) to give the title compound (694 mg, 84 %) as a colorless oil. ^{1}H NMR (400 MHz, CHLOROFORM-*d*) δ ppm 9.80 (1 H, s) 5.82 (2 H, s) 4.79 (2 H, s) 3.51 - 3.58 (2 H, m) 0.82 - 0.90 (11 H, m) 0.08 (6 H, s) -0.05 (9 H, s). LCMS: *m/z* 405 (M+1).



37

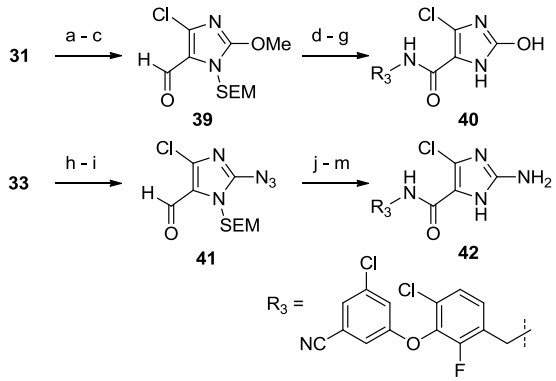
Compound 37. 2-(((tert-butyldimethylsilyl)oxy)methyl)-4-chloro-1-((2-(trimethylsilyl)ethoxy)methyl)-1H-imidazole-5-carboxylic acid. A solution of $NaClO_2$ (1.55 g, 17 mmol) and $NaH_2PO_4 \cdot H_2O$ (1.42 g, 10.3 mmol) in H_2O was added to a mixture of 2-(((tert-butyldimethylsilyl)oxy)methyl)-4-chloro-1-((2-(trimethylsilyl)ethoxy)methyl)-1H-imidazole-5-carbaldehyde (**36**) (694 mg, 1.7 mmol), 2-methy-2-butene (10.8 mL, 21.5 mmol, 2 M in THF), and tBuOH (1.33 mL) in THF (5.5 mL) at room temperature. The mixture was stirred overnight and separated and the aqueous layer was extracted with EtOAc. The combined extracts were dried over sodium sulfate and concentrated. The residue was purified by column chromatography (0-80 % 0.1 % formic acid solution of EtOAc in DCM) to give the title compound (740 mg, > 99 %) as a white solid. ^{1}H NMR (400 MHz, CHLOROFORM-*d*) δ ppm 5.83 (2 H, s) 4.80 (2 H, s) 3.56 (2 H, t, *J*=8.1 Hz) 0.83 - 0.91 (11 H, m) 0.08 (6 H, s) -0.04 (9 H, s). LCMS: *m/z* 421 (M+1).



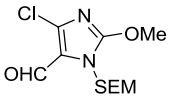
38

Compound 38. 4-Chloro-*N*-({4-chloro-3-[(3-chloro-5-cyanophenyl)oxy]-2-fluorophenyl}methyl)-2-(hydroxymethyl)-1*H*-imidazole-5-carboxamide. The reaction procedure and purification process are identical to that outlined herein except that 4-chloro-2-({[(1,1-dimethylethyl)(dimethyl)silyl]oxy}methyl)-1-({[2-(trimethylsilyl)ethyl]oxy}methyl)-1*H*-imidazole-5-carboxylic acid (**37**) (373 mg, 1.75 mmol) was used. The title compound (458 mg, 56 %) was obtained as a white solid. ^{1}H NMR (400 MHz, DMSO-d_6) δ ppm 8.29 (1 H, br. s.) 7.80 (1 H, s) 7.42 - 7.52 (3 H, m) 7.36 (1 H, br. s.) 5.51 (1 H, t, *J*=5.6 Hz) 4.49 (2 H, br. s.) 4.39 (2 H, d, *J*=5.6 Hz). LCMS: *m/z* 469 (M+1).

Synthesis of 2-OH and 2-NH_2 substituted imidazoles 40 and 42. Once again 2-bromo-4,5-dichloro-imidazole **31** proved to be a useful starting material as the bromide could be displaced with NaOMe in the presence of CuBr and subsequently converted to aldehyde **39** (see scheme below). After oxidation to the corresponding acid and coupling to benzylamine **6**, the *O*-methyl group was removed with BBr_3 to give the 2-OH analog **40** (Table 2). The 2-NH_2 derivative **42** was prepared via a similar approach. In this case, the methylsulfide intermediate **33** was oxidized to the sulfone with mCPBA and displaced with NaN_3 to give the corresponding aldehyde **41** in 69% yield. Upon oxidation of the aldehyde and removal of the SEM group, the acid was coupled to benzylamine **6**. Lindlar reduction of the azide provided the amino analog **42** in good yield.

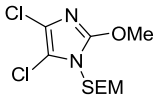


Reagents and conditions: Synthesis of **40**: (a) NaOMe, CuBr (67%); (b) n-BuLi, THF, -78 °C; (c) DMF (80%); (d) $NaClO_2$, $NaH_2PO_4.H_2O$, 2-Me-2-butene, *t*BuOH, THF (quant.); (e) EDC, HOBT, **6**, DMF; (f) TFA, DCM (56% over 2 steps); (g) BBr_3, DCM (15%). Synthesis of **42**: (h) mCPBA; (i) NaN_3, DMF (69% over 2 steps); (j) $NaClO_2$, $NaH_2PO_4.H_2O$, 2-Me-2-butene, *t*BuOH, THF (quant.); (k) TFA, DCM; (l) EDC, HOBT, **6**, DMF; (m) Lindlar catalyst, H_2 (50 psi), EtOAc (53% from **41**).



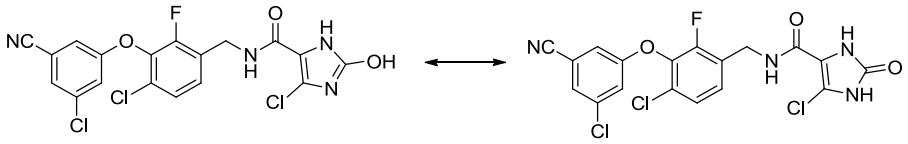
39

Compound 39. 4-chloro-2-(methyloxy)-1-({[2-(trimethylsilyl)ethyl]oxy}methyl)-1H-imidazole-5-carbaldehyde.



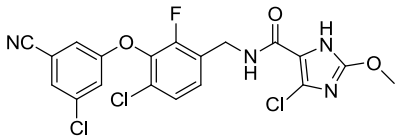
Step A: 4,5-dichloro-2-(methyloxy)-1-({[2-(trimethylsilyl)ethyl]oxy}methyl)-1H-imidazole. A solution of 2-bromo-4,5-dichloro-1-({[2-(trimethylsilyl)ethyl]oxy}methyl)-1H-imidazole (0.12 g, 0.35 mmol), sodium methoxide (0.12 mL of a 25% solution in methanol, 0.53 mmol) and CuBr (0.010 g, 0.07 mmol) in methanol (0.5 mL) was heated at 100 °C for 6h. Dilute aqueous NaCN and CH_2Cl_2 were added and the solution was stirred for a few minutes. The solution was extracted with EtOAc. The organic phase was dried (Na_2SO_4), filtered, and purified by silica gel chromatography (0-40% EtOAc/hex) to afford the title compound (0.070 g, 67%) as a clear oil. ^{1}H NMR (400 MHz, CHLOROFORM-*d*) δ ppm 5.11 (s, 2 H) 4.02 (s, 3 H) 3.51 - 3.63 (m, 2 H) 0.88 - 0.97 (m, 2 H) -0.01 (s, 9 H).

Step B. 4-chloro-2-(methyloxy)-1-({[2-(trimethylsilyl)ethyl]oxy}methyl)-1H-imidazole-5-carbaldehyde. The title compound **39** (0.42 g, 80%) was obtained as a clear oil from 4,5-dichloro-2-(methyloxy)-1-({[2-(trimethylsilyl)ethyl]oxy}methyl)-1H-imidazole (0.54 g, 1.82 mmol) using a procedure and process similar to that described herein. ^{1}H NMR (400 MHz, CHLOROFORM-*d*) δ ppm 9.68 (s, 1 H) 5.53 (s, 2 H) 4.15 (s, 3 H) 3.57 - 3.65 (m, 2 H) 0.87 - 0.96 (m, 2 H) -0.01 (s, 9 H).



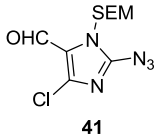
40

Compound 40. 5-chloro-N-({4-chloro-3-[(3-chloro-5-cyanophenyl)oxy]-2-fluorophenyl}methyl)-2-oxo-2,3-dihydro-1H-imidazole-4-carboxamide.



Step A: 4-chloro-N-({4-chloro-3-[(3-chloro-5-cyanophenyl)oxy]-2-fluorophenyl}methyl)-2-(methyloxy)-1H-imidazole-5-carboxamide. The procedure and process are similar to that described herein except that 4-chloro-2-(methyloxy)-1-({[2-(trimethylsilyl)ethyl]oxy}methyl)-1H-imidazole-5-carbaldehyde (0.42 g, 1.46 mmol) was employed to provide the acid as a clear oil which was used without further purification. The above acid (0.60 mmol) and 3-{[3-(aminomethyl)-6-chloro-2-fluorophenyl]oxy}-5-chlorobenzonitrile (0.19 g, 0.60 mmol) were employed in a similar process described herein to prepare the title compound (0.16 g, 56%) as a white solid after deprotection and purification by Reverse-Phase HPLC (water:acetonitrile with 0.1% TFA). ^{1}H NMR (400 MHz, CHLOROFORM-*d*) δ ppm 9.67 (br. s., 1 H) 7.28 - 7.40 (m, 3 H) 7.17 (s, 1 H) 7.03 (s, 1 H) 6.99 (t, J=5.77 Hz, 1 H) 4.69 (d, J=5.91 Hz, 2 H) 4.04 (s, 3 H). ES-LCMS: m/z 469.0, 471.0 (M+1).

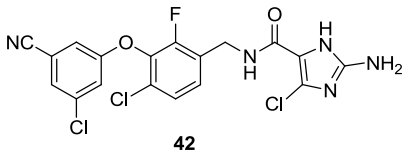
Step B. 5-chloro-N-({4-chloro-3-[(3-chloro-5-cyanophenyl)oxy]-2-fluorophenyl}methyl)-2-oxo-2,3-dihydro-1H-imidazole-4-carboxamide (40). BBr_3 (0.28 mL of a 1M solution in CH_2Cl_2, 0.28 mmol) was added to a solution of 4-chloro-N-({4-chloro-3-[(3-chloro-5-cyanophenyl)oxy]-2-fluorophenyl}methyl)-2-(methyloxy)-1H-imidazole-5-carboxamide (0.044 g, 0.093 mmol) in CH_2Cl_2 (1 mL). After 2h, additional BBr_3 (0.30 mL of a 1M solution in CH_2Cl_2, 0.30 mmol) was added and the reaction mixture was stirred for 3 days. The solution was evaporated, additional BBr_3 (1 mL of a 1M solution in CH_2Cl_2, 1 mmol) was added and the reaction mixture was stirred for 5 days. Sat'd $NaHCO_3$ was added and the mixture was extracted with EtOAc and then CH_2Cl_2. The organic phase was dried (Na_2SO_4), filtered, and purified by Reverse-Phase HPLC (water:acetonitrile with 0.1% TFA) to provide the title compound (0.006 g, 15%) as a white solid. ^{1}H NMR (400 MHz, DMSO-d_6) δ ppm 11.38 (s, 1 H) 10.41 (s, 1 H) 8.06 (t, J=5.68 Hz, 1 H) 7.82 (s, 1 H) 7.48 - 7.54 (m, 2 H) 7.46 (t, J=1.97 Hz, 1H) 7.38 (t, J=8.00 Hz, 1 H) 4.46 (d, J=5.57 Hz, 2 H). ES-LCMS: m/z 454.9, 456.9 (M+1).



41

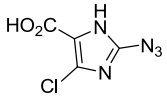
Compound 41. 2-azido-4-chloro-1-((2-(trimethylsilyl)ethoxy)methyl)-1H-imidazole-5-carbaldehyde. mCPBA (23.26 g, 94 mmol) was added to a solution of 4-chloro-2-(methylthio)-1-({[2-(trimethylsilyl)ethyl]oxy}methyl)-1H-imidazole-5-carbaldehyde (**33**) (12.59 g, 41.0 mmol) in dichloromethane (350 ml) at 0 °C. After 1h, the solution was removed from the ice bath and stirred at

room temperature for 4 hours. The solid was removed by filtration and sat'd $NaHCO_3$ was added to the filtrate and it was extracted with additional dichloromethane. The organic phase was dried (Na_2SO_4), filtered and concentrated to afford the sulfone as an off-white solid which was used without further purification. A solution of the sulfone and sodium azide (4.00g, 61.5 mmol) in DMF (200 mL) was stirred for 1h. Sat'd $NaHCO_3$ was added while cooling in an ice bath and the reaction mixture was extracted with CH_2Cl_2 (3 x 100 mL). The organic layer was dried (Na_2SO_4), filtered, concentrated, and purified by silica gel chromatography (0-30% EtOAc/hexanes) to provide the title compound as a yellow oil (8.53 g, 69 %). ^{1}H NMR (400 MHz, CHLOROFORM-*d*) δ ppm 9.73 (s, 1 H), 5.52 (s, 2 H), 3.55 - 3.67 (m, 2 H), 0.87 - 0.98 (m, 2 H), 0.00 (s, 9 H).



42

Compound 42. 2-amino-4-chloro-N-({4-chloro-3-[(3-chloro-5-cyanophenyl)oxy]-2-fluorophenyl}methyl)-1H-imidazole-5-carboxamide.

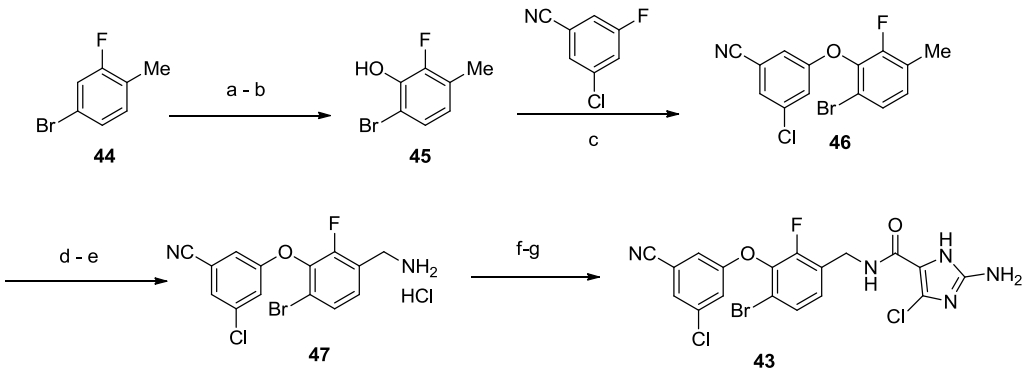


Step A. 2-azido-4-chloro-1H-imidazole-5-carboxylic acid. A solution of $NaClO_2$ (21.7 g, 240 mmol) and $NaH_2PO_4.2H_2O$ (19.9 g, 144 mmol) in H_2O (51 mL) was added to a stirred solution of 2-azido-4-chloro-1-({[2-(trimethylsilyl)ethyl]oxy}methyl)-1*H*-imidazole-5-carbaldehyde (**41**) (7.2 g, 24.0 mmol) and 2-methyl-2-butene (144 mL of a 2M solution in THF, 2887 mmol) in THF (72 mL) and t-BuOH (17 mL). The reaction mixture was stirred at room temperature for 1 hour and extracted with EtOAc. The organic layer was dried (Na_2SO_4), filtered and dried to provide 2-azido-4-chloro-1-({[2-(trimethylsilyl)ethyl]oxy}methyl)-1*H*-imidazole-5-carboxylic acid as a clear oil that was assumed to be quantitative and used without further purification. The above acid (1.40 mmol) was stirred in a solution of TFA (2.15 mL, 27.9 mmol) in CH_2Cl_2 (2 mL) for 1 hour at room temperature. Saturated $NaHCO_3$ was added and the mixture was washed with EtOAc. The aqueous layer was isolated, acidified with 1N HCl and extracted thoroughly with EtOAc. The organic layer was dried (Na_2SO_4), filtered, concentrated, and triturated with hexanes and again with diethyl ether to provide the title compound (assumed quant.) as a yellow solid. ^{1}H NMR (400 MHz, DMSO-d_6) δ ppm 13.33 (br. s.,1 H), 12.77 (br. s., 1 H).

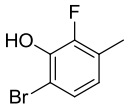
Step B. 2-amino-4-chloro-N-({4-chloro-3-[(3-chloro-5-cyanophenyl)oxy]-2-fluorophenyl}methyl)-1H-imidazole-5-carboxamide (42). A solution of 2-azido-4-chloro-1H-imidazole-5-carboxylic acid (0.26 g, 1.4 mmol), 3-{[3-(aminomethyl)-6-chloro-2-fluorophenyl]oxy}-5-chlorobenzonitrile (0.44 g, 1.40 mmol), 1-Ethyl-3-(3-dimethyllaminopropyl)carbodiimide hydrochloride (0.32 g, 1.68 mmol) and N-hydroxybenzotriazole (0.28 g, 1.82 mmol) was stirred at room temperature overnight. $NaHCO_3$ was added and the solution was extracted with 9:1 CH_2Cl_2:MeOH. The combined organic phase was dried (Na_2SO_4), filtered, and concentrated to give 2-azido-4-chloro-*N*-({4-chloro-3-[(3-chloro-5-cyanophenyl)oxy]-2-fluorophenyl}methyl)-1*H*-imidazole-5-carboxamide which was used without further purification after extraction. A solution of the above crude azide and catalytic $Pd/CaCO_3$ (poisoned with lead) in EtOAc (20 mL) was then stirred under 50 psi hydrogen gas for 2h. The solution was filtered through celite, evaporated and purified by silica gel chromatography (0-10% MeOH/CH_2Cl_2) to afford the title compound (0.33 g, 53% from **41**) as a white solid. ^{1}H NMR (400 MHz, DMSO-d_6) δ ppm 10.95 (s, 1

H), 7.82 (s, 1 H), 7.77 (t, J=5.86 Hz, 1 H), 7.48 - 7.54 (m, 2 H), 7.46 (t, J=2.01 Hz, 1 H), 7.34 (t, J=7.97 Hz, 1 H), 5.82 (s, 2 H), 4.49 (d, J=5.77 Hz, 2 H). ES-LCMS: m/z 454.0, 456.0, (M+1).

Compound 43. 2-amino-*N*-({4-bromo-3-[(3-chloro-5-cyanophenyl)oxy]-2-fluorophenyl}methyl)-4-chloro-1*H*-imidazole-5-carboxamide.

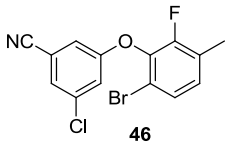


Reagents and conditions: (a) LDA, $B(OMe)_3$, THF, -75°C; (b) H_2O_2 (89% from 44); (c) K_2CO_3, DMA, 150 °C (64%); (d) NBS, $(BzO)_2$, DCE; (e) NH_3, MeOH, then HCl (56% from 46); (f) EDC, HOBT, 2-azido-4-chloro-1H-imidazole-5-carboxylic acid, DMF; (g) Lindlar cat., 50 psi H_2, EtOAc (44% from 47).



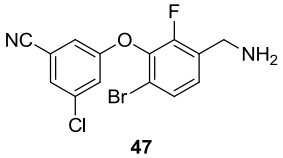
45

Compound 45. 6-bromo-2-fluoro-3-methylphenol. A 3L three neck round bottom flask, equipped with overhead stirrer, was charged with 2M LDA in hept/THF/ethylbenzene (300 mL, 600 mmol) and cooled to -75 °C. A solution of 4-bromo-2-fluoro-1-methylbenzene, **44**, (100g, 529 mmol) in THF (700 mL) was added dropwise over 35 min maintaining the internal temp below -67 °C. The resulting solution was stirred at -70 °C for ca. 2 hours. Trimethyl borate (66 mL) was added dropwise and stirring was continued for 20 min at -70 °C. Additional 2M LDA (100 mL, 200 mmol) was added dropwise followed by more trimethyl borate (20 mL, 179 mmol) and stirring was continued at -70°C for 1 hr and the mixture was allowed to warm to 0 °C. A solution of glacial acetic acid (110 mL) in water (750 mL) was rapidly added with good stirring. Upon completion of addition, 30% H_2O_2 (130 mL) was added dropwise over 20 min. The solution was stirred at room temperature for 1.5 hours. The layers were separated and the aqueous phase was adjusted to pH ~ 6 with 1N HCl and extracted once with EtOAc (600 mL). The combined THF and EtOAc layers were washed with 10% Na_2SO_3, and 1N HCl and then extracted with 1N NaOH (3 x 600 mL). The combined NaOH layers were acidified with 6 N HCl (300 mL) and extracted with DCM (2 x 500 mL). The combined DCM layers were dried over $MgSO_4$, filtered and concentrated to afford the title compound (97g, 89%) as a light amber oil which partially crystallized on standing. ^{1}H NMR (400 MHz, DMSO-d_6) δ ppm 2.13 (d, *J*=2.2 Hz, 3 H), 6.62 (t, *J*=7.8 Hz, 1 H), 7.14 (dd, *J*=8.2, 0.5 Hz, 1 H), 10.16 (br. s., 1 H).



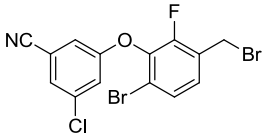
46

Compound 46. 3-(6-bromo-2-fluoro-3-methylphenoxy)-5-chlorobenzonitrile. A stirred mixture of 3-chloro-5-fluorobenzonitrile (106 g, 681 mmol), 6-bromo-2-fluoro-3-methylphenol (154 g, 750 mmol) and potassium carbonate (188 g, 1363 mmol) in N,N-dimethylacetamide (DMA) (1.1 L) was heated at 150°C for 7 hours. The reaction was allowed to come to ambient temperature as it stirred overnight. The mixture was diluted with isopropanol (1.1L) and water (5.5L) was added over 45 minutes. The reaction was stirred for 4 hours. The resulting solid was filtered, rinsed with water (1L), isopropanol/water (1:3, 1.2L) and air dried for 30 minutes. The solid was slurried in isopropanol (600mL) and stirred for 30 minutes. Water was added (600mL) and the solution was stirred overnight. The resulting solid was filtered, rinsed with 50% isopropanol/water (400mL), air dried for 5 hours and then in a vacuum oven at 60 °C for 18 hours to afford the desired product (147.3g, 64%) as a tan solid. ^{1}H NMR (400 MHz, CHLOROFORM-*d*) δ ppm 7.29 - 7.35 (m, 2 H), 7.13 (s, 1 H), 7.03 (t, *J*=7.8 Hz, 1 H), 6.97 (d, *J*=0.7 Hz, 1 H), 2.28 (d, *J*=1.8 Hz, 3 H).



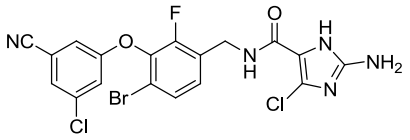
47

Compound 47. 3-(3-(aminomethyl)-6-bromo-2-fluorophenoxy)-5-chlorobenzonitrile.



Step A. 3-(6-bromo-3-(bromomethyl)-2-fluorophenoxy)-5-chlorobenzonitrile. To a solution of 3-[(6-bromo-2-fluoro-3-methylphenyl)oxy]-5-chlorobenzonitrile (144 g, 423 mmol) in 1,2-dichloroethane (DCE) (1.4 L) was added N-bromosuccinimide (90 g, 507 mmol) and benzoyl peroxide (2.73 g, 8.45 mmol). The reaction mixture was heated at reflux for 20 hours, washed with 1N NaOH (2x 1L), brine (1L), dried over magnesium sulfate and concentrated down to afford the desired crude product (188g) as an amber oil. NMR showed the desired product (70%), dibromo product (10%), starting material (20%) and DCE. The material was used without further purification. ^{1}H NMR (400 MHz, CHLOROFORM-*d*) δ ppm 7.44 (dd, *J*=8.4, 1.5 Hz, 1 H), 7.29 - 7.35 (m, 1 H), 7.25 (t, *J*=7.8 Hz, 1 H), 7.13 (d, *J*=1.9 Hz, 1 H), 6.98 (s, 1 H), 4.45 (s, 2 H).

Step B. 3-(3-(aminomethyl)-6-bromo-2-fluorophenoxy)-5-chlorobenzonitrile (47). A solution of 7M ammonia (2.402 L, 1680 mmol) in methanol was cooled to 10ºC and a solution of crude 3-{[6-bromo-3-(bromomethyl)-2-fluorophenyl]oxy}-5-chlorobenzonitrile (188 g, 314 mmol) in dichloromethane (200mL) was added over 6 hours. The reaction mixture was allowed to warm to ambient temperature and stirred overnight. The reaction was concentrated down, partitioned in ethyl acetate (1.5L), water (1.5L), 5% Na_2CO_3 (1.5L) and stirred for 1 hour. The layers were separated and the ethyl acetate layer was dried over magnesium sulfate and concentrated. The residue was taken up in ethyl acetate (1L), treated with HCl (4M in 1,4-dioxane, 65mL) and stirred for 30 minutes. The resulting solid was filtered, rinsed with ether and dried to afford the desired product HCl salt (92.6g, 56%, over 2 steps) as an off-white solid. ^{1}H NMR (400 MHz, *DMSO*-d_6) δ ppm 8.59 (br. s., 4 H), 7.80 (t, *J*=1.49 Hz, 1 H), 7.71 (dd, *J*=8.48, 1.18 Hz, 1 H), 7.55 (t, *J*=7.81 Hz, 1 H), 7.42 -7.46 (m, 2 H), 4.05 (s, 2 H).



43

Compound 43. 2-amino-*N*-({4-bromo-3-[(3-chloro-5-cyanophenyl)oxy]-2-fluorophenyl}methyl)-4-chloro-1*H*-imidazole-5-carboxamide. EDC (0.331 g, 1.726 mmol) and HOBT (0.233 g, 1.726 mmol) were added to a solution of 3-{[3-(aminomethyl)-6-bromo-2-fluorophenyl]oxy}-5-chlorobenzonitrile (0.558 g, 1.569 mmol) and 2-azido-4-chloro-1H-imidazole-5-carboxylic acid (0.294 g, 1.569 mmol) in DMF (7 mL). The mixture was stirred at room temperature overnight. The reaction mixture was diluted with EtOAc and washed with water and brine and the organic layer was dried over sodium sulfate and concentrated to give the crude title compound (0.850 g) as a light yellow solid. This crude product was taken on to the next step without further purification.

Lindlar Catalyst (0.167 g, 0.078 mmol) was added to a suspension of 2-azido-N-({4-bromo-3-[(3-chloro-5-cyanophenyl)oxy]-2-fluorophenyl}methyl)-4-chloro-1H-imidazole-5-carboxamide (0.824 g, 1.569 mmol) in ethyl acetate (30 mL) in a pressure vessel. The vessel was evacuated and flushed with nitrogen, then evacuated and filled with hydrogen (50 psi). The reaction mixture was stirred for 7 h and filtered through celite. The filtrate was evaporated to dryness and the residue was triturated with ethanol and dried under vacuum to give the title compound (0.350 g, 44%) as a white solid. ^{1}H NMR (400 MHz, DMSO-d_6) δ ppm 10.95 (br. s., 1 H), 7.80 (s, 1 H), 7.76 (t, 1 H), 7.60 (d, 1 H), 7.48 (s, 1 H), 7.42 (m, 1 H), 7.26 (t, 1 H), 5.79 (br. s., 2 H), 4.45 (d, 2 H). ES MS m/z 498 (M-H).

II. FTIR analysis of compound 40.

Experimental details:

- Spectrometer: $CDCl_3$ solution spectra acquired using Bruker Vector 27, ATR-IR spectrum acquired using Bruker Vector 22 with Durascope ATR accessory
- Frequency Range: 4000-950 cm^{-1} ($CDCl_3$ solution-IR) and 4000-650 cm^{-1} (ATR-IR)
- Data acquired: $CDCl_3$ solution spectra and ATR-IR spectra of films
- Additional Processing: $CDCl_3$ solution spectra – solvent bands removed by subtraction, ATR-IR spectra of films – none

FTIR analysis: The IR spectrum acquired from a $CDCl_3$ solution of compound 40 is shown in Figure 1. Two carbonyl bands have been identified in the experimental spectrum. No O-H stretching band is observed. In Figure 2 below the ATR-IR spectrum of a film of compound 40 (black) is overlaid with the ATR-IR spectrum of a deuterated film of compound 40 (red). No O-H stretching band is observed. Two carbonyl bands are observed and identified in both spectra. These observations support no presence of the enol tautomer (O-H stretching band) in compound 40 under these experimental conditions.

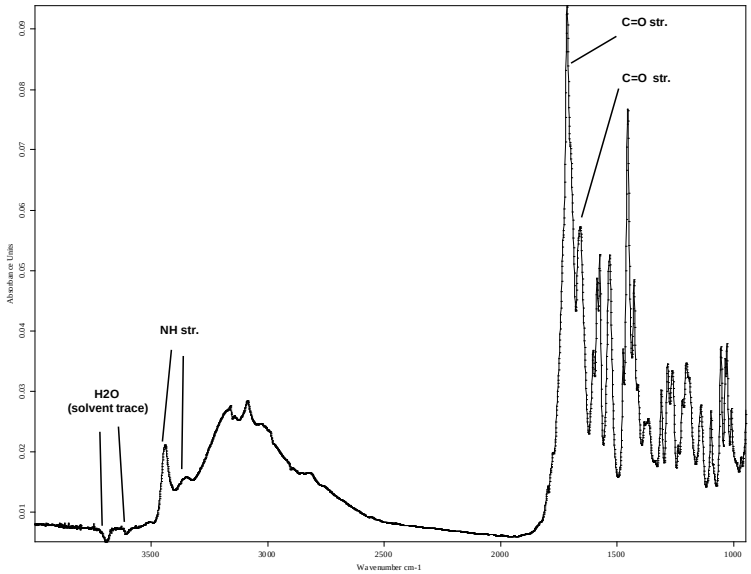


Figure 1. $CDCl_3$ solution IR spectrum of compound 40.

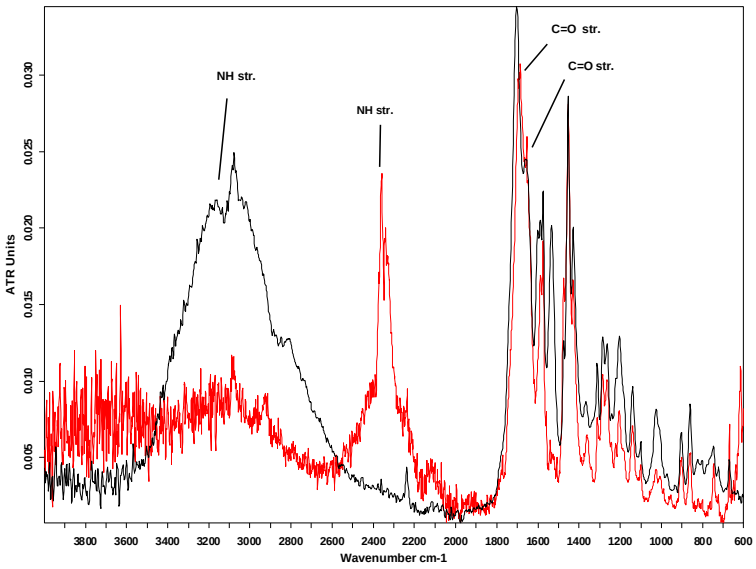


Figure 2. ATR-IR spectrum of a film of compound 40 (black) overlaid with ATR-IR spectrum of a deuterated film of compound 40 (red).

III. Biological Section - Inhibition of Viral Replication.

HeLa Cell Assay

The HeLa cell assay described herein is a modified version of Kimpton J. and Emerman M., Detection of replication-competent and pseudotyped human immunodeficiency virus with a sensitive cell line on the basis of activation of an integrated β -galactosidase gene, J. *Virol.* 66:2232-2239 (1992), in which HIV-1 infection is detected by the activation of an HIV-LTR driven β -galactosidase reporter that is integrated into the genome of a CD4+ HeLa cell line. Quantitation of β- galactosidase is achieved by measuring the activation of a chemiluminescent substrate (Applied Biosystems). The concentration of each compound required to inhibit 50% (EC50) of the HIV-1 induced β -galactosidase signal, relative to untreated controls, is determined for each isogenic, recombinant virus.

A. Materials

HeLa-CD4-LTR- β -gal cell line (AI DS Research and Reference Reagent
Program, Division of AIDS, NIAID)
DMEM (GibcoBRL # 12430-047)
Trypsin-EDTA (GibcoBRL #25300-054)
Heat inactivated Fetal Bovine Serum (FBS) (Hyclone # SH30070.03)
Geneticin (GibcoBRL # 10131-035)
Hygromycin B (GibcoBRL #1687-010)
96-well, black, clear-bottom, tissue culture-treated plates (Costar # 3904)
0.45 micron cellulose acetate filtration unit (Corning # 430768)
DEAE-dextran (Sigma # D-9885)
Phosphate Buffered Saline (PBS) (GibcoBRL #14190-144)
Dimethyl Sulfoxide (DMSO) (ATCC # 741625)
Gal-Screen Reporter Gene Assay System (Applied Biosystems # T1 031)
Human Serum Albumin (Sigma #A1653)
Human alpha-1 acid glycoprotein (Sigma #G9885)

B. Growth and Maintenance of the CD4-HIV LTR-β-gal HeLa cell line.

HeLa-CD4-LTR- β -gal cells are propagated in DMEM containing 10% fetal bovine serum + 0.2 mg/ml geneticin + 0.1 mg/ml hygromycin B. Cells are split by standard trypsinization when confluency reaches 80% (roughly every 2 to 3 days).

C. Construction of HIV-1 reverse transcriptase (RT) mutants.

DNA encoding the HIV-1 reverse transcriptase is subcloned from a M13 phage into a general shuttle vector, pBCSK+, as a ~1.65 kbp EcoRI/Hindlll ended DNA fragment. The HIV DNA insert of the resulting plasmid is completely sequenced on both strands prior to use in site directed mutagenesis experiments. Specific amino acid replacements are made using Stratagene Quick Change reagents and mutagenic oligonucleotides. Following mutagenesis, the entire mutant RT coding sequence is verified by sequencing both DNA strands.

D. Construction of isogenic HIV-1 RT mutant virus

Mutant HIV-1 strains and wild type strains are isolated by a modified Recombinant Virus Assay (Kellam P. and Larder B. Recombinant virus assay: a rapid, phenotypic assay for assessment of drug susceptibility of human immunodeficiency virus type 1 isolates, *Antimicrobial Agents and Chemotherapy,* 38:23-30, 1994). Ten million MT4 T-cells (maintained in RPMI containing 10% fetal bovine serum, split 1:5 every 5 to 6 days) are co-transfected with EcoRI/Hindlli digested mutant RT plasmid and Bst Ell-digested HIV-

$1_{HxB2\Delta RT}$ DNA in the presence of DMRIE-C transfection reagent (Gibco) according to supplier's recommended protocol. Each mutant RT coding sequence is crossed into the RT-deleted HIV-1 viral DNA backbone by in vivo homologous recombination. Transfected cell cultures are expanded and monitored until syncitia formation and CPE are extensive. Virus is harvested by clear spin of the culture supernatants, filtration of the supernatants through a 0.45 micron membrane and frozen at – 80°C as primary stocks. Recombinant progeny virus is sequenced in the RT region to confirm the mutant genotype. Some virus stocks require further expansion by infection of MT4 cells, harvested as above and stored as frozen aliquots. All stocks are titered in HeLa MAGI cells for assay.

E. Titering of virus stocks.

HIV-1 virus stocks are titered in the HeLa-CD4-LTR- β-gal assay system to establish the appropriate infecting dose. The endpoint for this assay is relative light units (RLUs), and titer is recorded as RLUs/ml. Virus stocks are diluted (serial 1:2) into DMEM containing 10% FBS plus 25ug/ml DEAE-dextran and assayed as described in the "Experimental Protocol" section below without test compound.
A "multiplicity of infection" (MOl) defined as infectious units per cell is usually not calculated but is typically <<1.0.

F. Experimental Protocol

Day 1
1. Seed 96-well plate(s) (Costar #3904) with HeLa-CD4-LTR- β-gal @ 3 X 10^3 cells per well in 100ul DMEM containing 10% FBS. Incubate @ 37°C, 5% CO2 overnight.

Day 2
1. Thaw virus stock in a water bath (room temperature) and dilute into DMEM +10% FBS + 25ug/ml DEAE-dextran to an infectious dose of approximately 10 million RLU/ml. The dilution of virus will vary depending on the titer of the stock (see "Titering of virus stocks" above).
2. Remove all of the media from every well with an 8 or 12-channel manifold aspirator. Work with one plate at a time to prevent drying of the HeLa-CD4-LTR-β-gal monolayer. Add 35ul (approximately 350,000 total RLUs) of diluted virus to each well. Incubate @ 37°C, 5% CO2 for 2 hours.
3. During the virus adsorption period prepare compound titration plates at 1.35X final concentration. In general, test compounds are titrated robotically on a Beckman 2000 laboratory automation workstation (Beckman Coulter) in a four-fold stepwise manner from 2.7uM (2uM final) down to 0.01 nM (0.008nM final). This scheme will allow for 8 test compounds per 96-well plate with 10 dilution points and 2 controls per compound (n=1). Test compounds are titrated into DMEM + 10% FBS + 0.135% DMSO (0.1% final). The final volume of titrated compound in each well should be at least 150ul and DMSO should be at 0.135% (0.1% final) including the no compound controls.
4. With a RapidPlate *96/384* liquid handling system (Zymark) remove 100ul of titrated compound from every well of the titration plate prepared in step 3 above and add to the virus adsorption plate (step 2 above).
5. Incubate @ 37°C, 5% CO2 for 72 hours.

Day 5
1. With a RapidPlate *96/384* liquid handling system (Zymark) reduce supernatants to 50uL and add 50uL of reconstituted Gal-Screen according to manufacturer's recommended protocol.
2. Mix plate(s) vigorously on a platform shaker.
3. Read plate(s) in a Topcount luminometer (Packard) at 1slwell.

Protein binding assays are performed exactly as described above except the tissue culture medium is supplemented with human serum albumin and human alpha-1 acid glycoprotein at a final concentration of 40mg/mL and 1mg/mL, respectively.

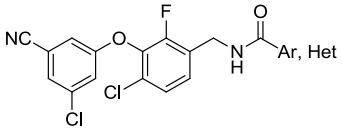
G. Data Analysis

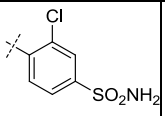
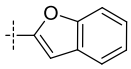
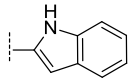
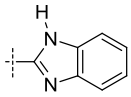
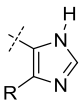
Raw data are transformed into percent of control by the following formula: (raw signal in each well / average raw signal for the two no compound controls in the same row)*100. Percent of control is plotted vs. compound concentration using either Robsage or Robofit programs (GSK). The default model is Y=Vmax*1-(x^n/(K^n+x^n)), however, any other model giving a reasonable estimation of the EC50 ("K" in formula) may be used.

The effect of protein binding is expressed as fold shift values and is calculated by dividing the EC50 of test compounds against wt virus in the HeLa assay in the presence of 40mg/mL human serum albumin plus 1mg/mL human alpha-1 acid glycoprotein by the EC50 of test compounds against wt virus under standard assay conditions. Protein adjusted EC50 values are calculated by multiplying the EC50 of test coumpounds derived against a mutant virus under standard assay conditions by the fold shift value.

IV. Antiviral data: standard deviations.

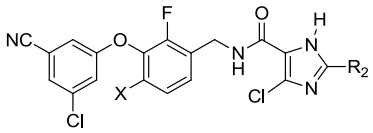
A. Table 1. Antiviral potency of aryl and heteroaryl analogs.



			EC_{50} ± SD (nM)			
Cmpd	**Ar, Het**	**R**	**WT**	**K103N**	**Y181C**	**Y188L**
EFV			0.51 ± 0.44 (n=137)	20 ± 17 (n=133)	1.0 ± 1.1 (n=121)	160 ± 170 (n=89)
ETR			0.6 ± 0.8 (n=60)	0.4 ± 0.6 (n=53)	2.4 ± 2.5 (n=39)	0.9 ± 1.0 (n=55)
TMC278			0.5 ± 0.4 (n=42)	0.5 ± 0.3 (n=39)	1.3 ± 0.8 (n=39)	3.7 ± 2.3 (n=39)
1			0.3 ± 0.3 (n=210)	0.5 ± 0.4 (n=194)	0.5 ± 0.4 (n=195)	18 ± 14 (n=160)
7			13 ± 6 (n=2)	60 ± 18 (n=2)	182 ± 17 (n=2)	>1000 (n=2)
8			52 ± 14 (n=2)	380 ± 310 (n=2)	340 (n=1)	>1000 (n=2)
9			8.9 ± 1.3 (n=3)	26 ± 9 (n=4)	58 ± 22 (n=4)	>1000 (n=4)
10			5.1 (n=1)	18 ± 1 (n=2)	27 ± 3 (n=2)	>1000 (n=2)
11			120 ± 20 (n=2)	460 ± 200 (n=2)	> 1000 (n=2)	>1000 (n=2)
12		H	9.7 ± 2.8 (n=2)	48 ± 19 (n=2)	260 ± 230 (n=2)	>1000 (n=2)
13		Me	2.4 ± 0.4 (n=2)	7.2 ± 5.2 (n=2)	15 ± 4 (n=2)	>1000 (n=2)
14		CF_3	0.14 ± 0.13 (n=2)	0.73 ± 0.28 (n=2)	1.4 ± 0.9 (n=2)	320 ± 40 (n=2)

15		Cl	0.1 ± 0.08 (n=19)	0.11 ± 0.08 (n=17)	0.2 ± 0.1 (n=17)	5.6 ± 3.2 (n=19)
16		Br	0.12 ± 0.07 (n=14)	0.13 ± 0.11 (n=14)	0.2 ± 0.3 (n=14)	7.1 ± 6.7 (n=16)

B. Table 2. Antiviral activity of imidazoles substituted at R_2.



			EC_{50} ± SD (nM)			
Cmpd	**X**	**R_2**	**WT**	**K103N**	**Y181C**	**Y188L**
15	Cl	H	0.1 ± 0.08 (n=19)	0.11 ± 0.08 (n=17)	0.2 ± 0.1 (n=17)	5.6 ± 3.2 (n=19)
28	Cl	Me	0.22 ± 0.26 (n=12)	0.17 ± 0.12 (n=11)	0.3 ± 0.13 (n=12)	11 ± 3 (n=12)
29	Cl	Et	0.28 ± 0.17 (n=15)	0.33 ± 0.21 (n=15)	0.77 ± 0.38 (n=15)	76 ± 23 (n=15)
30	Cl	nPr	0.25 ± 0.13 (n=2)	0.36 ± 0.39 (n=2)	1.3 ± 0.3 (n=2)	240 ± 170 (n=2)
38	Cl	CH_2OH	0.27 ± 0.01 (n=2)	0.32 ± 0.08 (n=2)	1.2 ± 0.9 (n=3)	24 ± 16 (n=3)
40	Cl	OH	5.3 (n=1)	14 (n=1)	57 (n=1)	>1000 (n=1)
42	Cl	NH_2	0.06 ± 0.05 (n=12)	0.06 ± 0.06 (n=10)	0.11 ± 0.08 (n=11)	0.94 ± 0.66 (n=13)
43	Br	NH_2	0.09 ± 0.08 (n=35)	0.1 ± 0.06 (n=29)	0.12 ± 0.08 (n=32)	0.5 ± 0.4 (n=36)

V. Protein adjusted antiviral data: standard deviation.

A. Antiviral activity of compound **43**, efavirenz, etravirine, and rilpivirine (TMC278)

	Compound 43			Efavirenz			Etravirine			Rilpivirine		
Virus	**n**	**EC50 [nM]**	**Std Dev**	**n**	**EC50 [nM]**	**Std Dev**	**n**	**EC50 [nM]**	**Std Dev**	**n**	**EC50 [nM]**	**Std Dev**
WTRVA	35	0.09	0.08	137	0.5	0.4	60	0.6	0.8	42	0.5	0.4
V179F	3	0.002	0.000	2	0.006	0.004	4	0.01	0.02	3	0.006	0.003
V179F/Y181I	2	0.03	0.01	1	0.03		2	169	5	2	19	11
E138G	3	0.20	0.04	2	0.4	0.1	4	0.7	0.3	3	0.9	0.3
V90I	3	0.09	0.06	2	0.4	0.2	4	0.2	0.1	3	0.4	0.1
H221Y	3	0.08	0.07	2	0.7	0.2	4	0.48	0.02	3	0.7	0.2
P236L	4	0.05	0.03	13	0.8	0.6	3	0.5	0.3	8	0.4	0.2
Y181I	3	0.18	0.05	2	0.9	0.0	4	29	4	3	38	19
V106A	30	0.08	0.07	126	1.0	0.9	52	0.4	0.8	39	0.4	0.3
Y181C	32	0.12	0.08	121	1.0	1.1	54	2.4	2.5	39	1.3	0.8
V106I	5	0.09	0.06	17	1.0	0.7	6	0.7	0.5	9	0.4	0.2
G190E	4	0.03	0.01	9	1.1	0.5	3	0.6	0.1	8	0.5	0.1
Y181V	3	0.24	0.11	2	1.3	0.8	4	50	31	3	35	9
Y188C	4	0.06	0.12	18	1.4	0.6	4	0.1	0.1	8	0.1	0.1
E138K	5	0.12	0.05	17	1.6	0.9	5	1.6	0.6	9	1.1	0.6
P225H	5	0.04	0.02	13	1.6	1.0	5	0.3	0.1	9	0.2	0.1
V106A/Y181C	5	0.06	0.02	19	2.0	0.9	5	2.0	1.0	9	1.0	0.3
V108I	5	0.04	0.02	16	2.2	1.3	6	0.5	0.6	9	0.2	0.1
V108I/Y181C	5	0.07	0.04	13	3.1	1.5	5	1.6	0.5	9	1.0	0.3
K101E	3	0.25	0.08	16	3.1	1.5	6	1.5	0.4	10	1.6	0.9
V106I/Y181C	5	0.40	0.18	21	4.2	2.7	6	5.9	4.1	9	3.6	1.1
V179F/Y181C	3	0.04	0.05	2	4.8	1.2	4	183	62	3	11	2
G190A	5	0.04	0.02	14	6.3	3.2	6	0.3	0.2	9	0.2	0.1
V106M	4	0.03	0.02	8	7.5	2.2	3	0.4	0.2	8	0.2	0.1
Y181C/F227C	3	3.2	1.3	2	10	2	4	36	14	3	23	7
K103N	29	0.1	0.06	133	20	17	53	0.4	0.6	39	0.5	0.3
L100I	5	0.02	0.01	23	25	14	6	1.2	0.9	9	0.4	0.4
K103N/Y181C	5	0.08	0.03	26	48	16	6	5.1	1.5	9	2.4	1.5
G190S	5	0.02	0.02	5	94	35	5	0.1	0.1	9	0.08	0.03
K103N/V108I	4	0.04	0.02	20	118	65	5	0.3	0.2	9	0.5	0.2
Y188L	36	0.50	0.35	89	158	168	55	0.9	1.0	39	3.7	2.3
K103N/P225H	5	0.06	0.06	23	162	91	6	0.4	0.2	9	0.5	0.5
K103N/Y181C/G190A	3	0.14	0.13	2	> 500		4	3.0	1.5	3	1.7	0.5
K103N/G190A	4	0.05	0.02	22	619	337	7	0.2	0.3	9	0.2	0.2
K103N/L100I	5	0.03	0.02	25	1422	650	7	4.4	1.8	9	4.5	1.8

B. Protein shift data for compound **43**, efavirenz, etravrine, and rilpivirine (TMC278).

The protein-adjusted EC_{50} (PA-EC_{50}) versus wild-type HIV-1 was measured in the presence of 40mg/mL human serum albumin (HSA) and 1mg/mL human alpha-1 acid glycoprotein (AAG) as described in the Biological Section. The fold-shift is the ratio of PA-EC_{50} / EC_{50}.

Compound	***Mean Fold Shift +/- SD**	**N**
Compound 43	71 +/- 53	29
efavirenz	23 +/- 15	23
etravirine	13 +/- 11	29
rilpivirine	116 +/- 68	31

C. Figure 6. Protein-adjusted EC_{50} (PA-EC_{50}) for compound **43**, efavirenz, etravirine, and rilpivirine (TMC278).

The PA-EC_{50} values for compound **43**, efavirenz, etravirine, and rilpivirine (TMC278) versus the panel of NNRTI resistant viruses were calculated from the EC_{50} (data highlighted above in Section V-A) multiplied by the protein fold-shift (data highlighted in Section V-B).

	PA-EC50			
Virus	**Compound 43**	**Etravirine**	**Rilpivirine**	**Efavirenz**
WT	7	8	61	12
V179F	0	0	1	0
V179F/Y181I	2	2274	2230	1
E138G	14	10	98	8
V90I	6	3	43	9
H221Y	6	6	82	15
P236L	3	7	44	17
Y181I	12	389	4354	20
V106A	6	6	47	22
Y181C	8	32	154	23
V106I	6	9	47	24
G190E	2	7	53	25
Y181V	17	675	3995	28
Y188C	4	1	8	32
E138K	9	21	127	36
P225H	3	3	22	37
V106A/Y181C	4	27	116	46
V108I	3	7	28	49
V108I/Y181C	5	22	115	69
K101E	18	21	188	71
V106I/Y181C	29	80	411	96
V179F/Y181C	3	2473	1302	109
G190A	3	4	21	143
V106M	2	5	26	169
Y181C/F227C	229	480	2614	223
K103N	7	6	53	463
L100I	1	16	46	570
K103N/Y181C	5	68	274	1088
G190S	1	2	10	2125
K103N/V108I	3	4	57	2675
Y188L	35	12	428	3566
K103N/P225H	4	6	53	3677
K103N/Y181C/G190A	10	41	196	11320
K103N/G190A	4	3	27	14007
K103N/L100I	2	59	515	32184

VI. In Vivo/In Vitro DMPK Profiling.

Pharmacokinetics

Male Sprague-Dawley rats (Charles River Labs, Raleigh, NC) were dosed intravenously at 1 mg/kg and orally at doses ranging from 5 mg/kg to 1000 mg/kg for pharmacokinetic evaluations. Blood samples were collected at various time points into EDTA tubes, and centrifuged to form plasma. Plasma samples were stored at -70°C until sample analysis. Samples were extracted by protein precipitation and analyzed by LC/MS/MS.

VII. Crystallography methods.

Protein Expression and Crystallography: Wild type and mutant (Y188L) p66 (1-560) subunits of RT (HXB-2 isolate) were expressed with nonviral residues MNS at the N-terminus in recombinant *E. coli* strain TG1. Wild type RT p51 (1-428) subunit was expressed with an N-terminal His-tag and TEV protease cleavage site (MAGHHHHHHGSAENLYFQ\G) in *E. coli* strain BL21[DE3]. RT heterodimers (wt or Y188L p66/6His-TEV-p51) were formed by lysing mixtures of cells (~2 g p66 cells:1g p51 cells) in 20 mM Na-HEPES, 500 mM NaCl, 1 mM $MgCl_2$, 12.5 mg/L AEBSF, 1 mM pepstatin A, 1 mM aprotinin, 5 mg/L each DNase and RNase, pH 8 at ~300 g cell paste/L of buffer by passing through a continuous flow pressure cell twice at 10,000 psi. The expression levels were such that these lysates had a molar excess of p51 to p66. Lysates were brought to and held at 37 °C with stirring for 30 minutes to enhance heterodimer formation. Following centrifugation and 0.2µ filtration, lysates were applied to a Nickel Sepharose FF (GE Healthcare) column equilibrated in 20 mM Na-Hepes, 500 mM NaCl, pH 7.5 (buffer A) and RT was eluted with an imidazole gradient (10CV 0-250 mM). RT heterodimer was pooled and the His-tag was removed with TEV protease (1 mg TEV:40 mg RT) during concentration with 30K MWCO Jumboseps (Pall) at 27 °C followed by overnight dialysis at 4 °C in Nickel buffer A. The cleaved RT was applied to the Nickel column and collected in the flowthrough and early gradient fractions. Cleaved heterodimer was pooled, concentrated, dialyzed in Q buffer A and applied to a Q-Sepharose HP (GE Healthcare) column equilibrated in 50 mM Tris-Cl, pH 8.5 (Q buffer A). Elution with a 7CV 0-110 mM NaCl gradient separated heterodimer from residual p51. RT heterodimer was exchanged into 10 mM Tris-Cl, 50 mM KCl, pH 8.5 and concentrated to 35-40 mg/mL with 30K Jumboseps and stored at -80 °C. Five molar excess compound was added to protein solution followed by 1 hour room temperature incubation. Hanging drops were set at 22 °C and cocrystals were grown in 100mM HEPES 7.5, 0.01M spermidine, and 1.1M potassium sodium tartrate. Prior to X-ray data collection, crystals were slow exchanged to 20% glycerol in mother liquor at 4 °C. X-ray data were collected at the Advance Photon Source, on beamlines 17-ID-B & 21-ID-F, and at the Canadian light source on beamline 08-ID-1. Structures were solved by standard crystallographic methods using programs from the both the CCP4[1] and Phenix[2] software suites. Molecular replacement was carried out using the program Phaser[3] from an internal RT structure originally solved using pdbcode 1HMV. Structures were refined using Refmac[4], and built using Coot[5]. Ligand parameters were generated using the elbow package[6].

Table 1: X-ray refinement statistics

Structure	RT Wild Type	RT Wild Type	RT Wild Type	RT Y188L
Compound #	15	38	43	43
Compound Name	GSK952	GSK500	GSK560	GSK560
Space Group	C2221	C2221	C2221	C2221
Cell (Å)	118.50 154.58 156.41	118.99 154.57 156.76	118.88 154.63 156.96	119.58 154.51 157..03
Data Resolution (Å)	40.0-2.49 (2.58-2.49)	50.0-2.90 (2.95 – 2.90)	80.0-2.12 (2.18 – 2.12)	50.0 - 2.35 (2.39 – 2.35)
Data Collection	CLS 08ID-1	APS 17ID	APS 21ID-F	APS 21ID-F
Detector	Mar 225	ADSC Q210	Mar225	Mar225
Rmerge %	6.3 (41.4)	9.1 (46.5)	6.3 (40.4)	6.4 (44.7)
I/sigmaI	30.9 (4.96)	21.8(3.7)	27.0 (4.7)	23.4 (4.15)
Completeness %	99.9 (100)	99.9 (100)	99.9 (100)	99.9 (100)
Average Redundancy	7.4 (7.4)	7.4 (6.8)	6.3 (6.2)	5.3 (5.4)
Unique Reflections	50530 (4976)	32529 (1600)	80302 (6609)	59250 (2904)
Wilson B	41.1	27.3	32.0	17.9
R-Factor %	20.37	20.85	19.74	19.84
Free R-Factor %	25.36	26..24	24.07	23.63
ProteinAtoms	7851	7869	7780	7754
Solvent Atoms	378	97	660	437
Ligand Atoms	28	30	29	29
Mean BFactor (Å**2)	39.11	20.47	28.94	38.10
RMS Bonded Bfactors (Å**2)	0.99	0.95	2.74	1.94
RMS Bond Lengths (Å)	0.007	0.007	0.010	0.011
RMS Bond Angles (˚)	1.053	1.114	1.28	1.35
RMS Torsion Angles (˚)	5.46	5.53	5.92	5.92
Molprobity				
Clash score	6.59 [98th%]	6.36 [100%]	5.77 [98%]	7.32 [98%]
Rotomer outliers	1.07	1.55%	0.84%	0.73%
Ramachandran outliers	0.21%	0.21%	0.31%	0.10%
Ramachandran favored	97.80%	96.24%	97.51%	97.38%
Cβ deviations	0	0	0	0
Bad bond Lengths	0	0	0	0
Bad bond Angles	0	0	0	0.10%
Overall Score	1.43 [100%]	1.75 [100%]	1.41 [99%]	1.52 [99%]
wwPdb Code	2YNI	2YNH	2YNG	2YNF

Numbers in parenthesis represent values for the high resolution shell; Numbers in square brackets are molprobity percentile scores

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