Supplemental Information

Discovery of Lirafugratinib (RLY-4008), a Highly Selective Irreversible Small-Molecule Inhibitor of FGFR2

Supplemental Figures

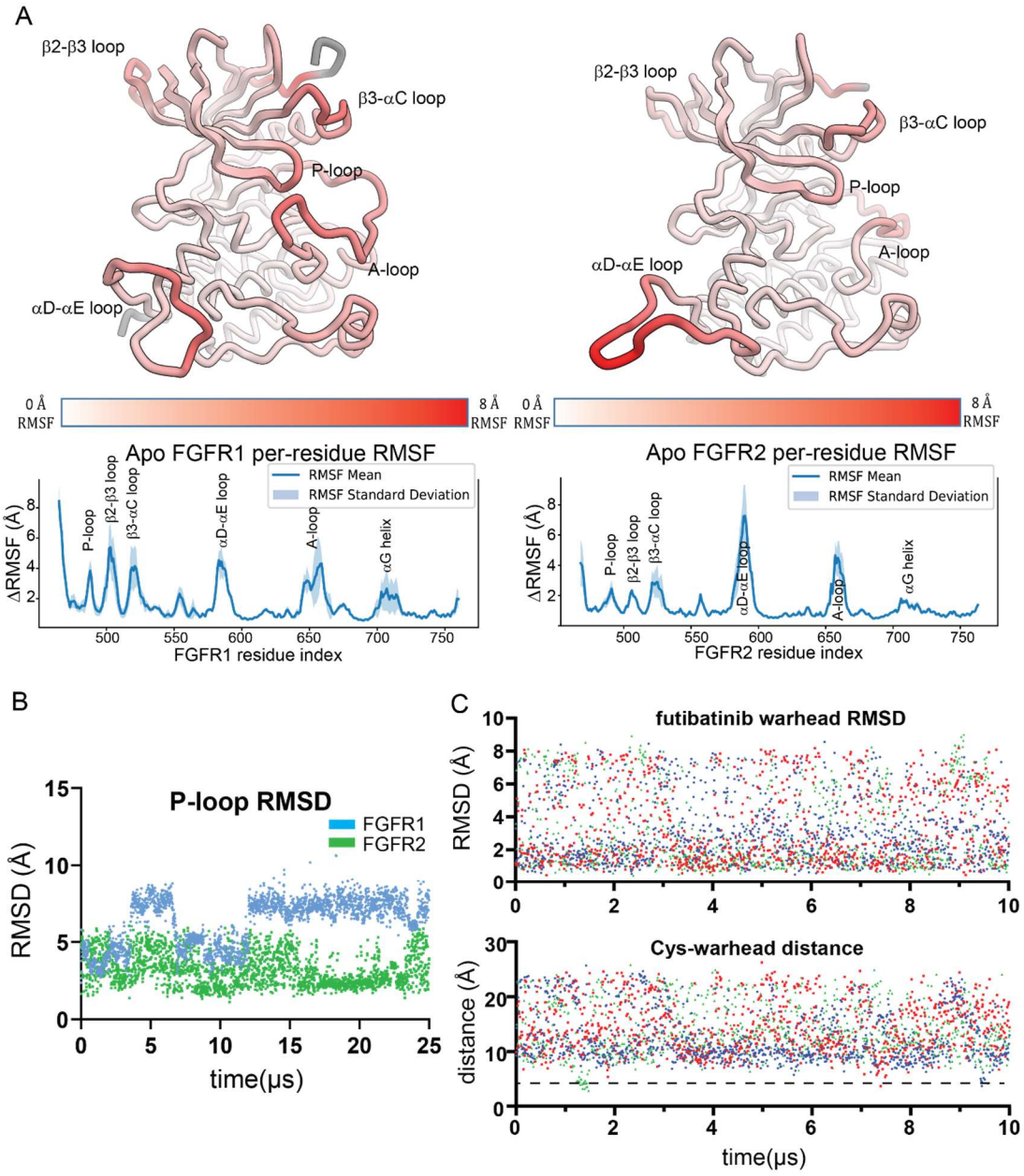


Figure S1: *The P-loop region has different dynamics in in FGFR1 and FGFR2* (A) RMSF values of the Cα atoms from apo simulations of FGFR1 (left) and FGFR2 (right) are plotted for

each residue (the line is a mean of three independent simulations, and the shading indicates the standard deviation of the three simulations). (B) P-loop RMSDs over time are plotted for apo simulations of FGFR1 and FGFR2. For the calculation, each trajectory was aligned on a set of N-lobe Cα atoms (FGFR1: residues 472–566; FGFR2: residues 475–569), and the RMSD was calculated using the Cα atoms of the P-loop residues (FGFR1: residues 484–492; FGFR2: residues 487–495) (C) The futibatinib warhead RMSD over time is plotted for three independent simulations (red, blue, and green; top). For the calculation, each trajectory was aligned on a set of N-lobe Cα atoms (residues 472–498, 509–517, 545–566, and 628–641), and RMSD with respect to the starting pose was calculated using futibatinib's pyrrolidine acrylamide moiety. Distance between the futibatinib warhead amide carbon atom and the Cys488 sulfur atom is plotted for three independent simulations (bottom).

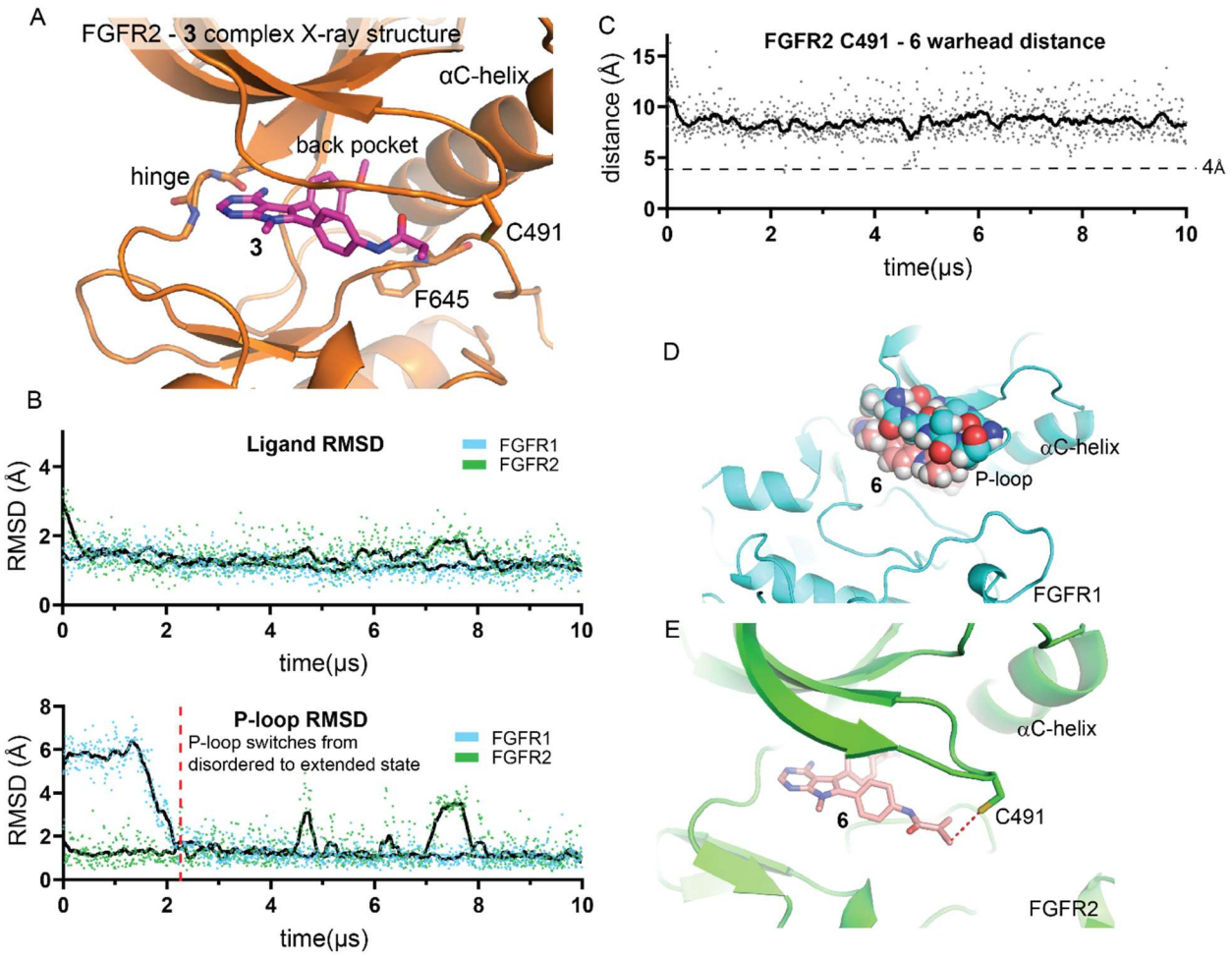


Figure S2*: Compound **6** adopts a stable binding pose in reversible simulations with FGFR2.* (A) X-ray structure of (3) bound with FGFR2. (B) RMSDs of **6** (top) and the P-loop (bottom) with respect to the last frame of each simulation are plotted for **6**-bound FGFR1 and FGFR2 simulations. For the ligand RMSD calculation, each trajectory was aligned on a set of N-lobe Cα atoms (FGFR1: residues 472–498, 509–517, 545–566, and 628–641; FGFR2: residues 475–501, 512–520, 548–569, and 631–644), and all ligand heavy atoms were used. For the P-loop RMSD calculation, each trajectory was aligned on a set of N-lobe Cα atoms (FGFR1: residues 472–566; FGFR2: residues 475–569), and the values were calculated using the Cα atoms of the P-loop residues (FGFR1: residues 484–492; FGFR2: residues 487–495). (C) Distance between the **6**

warhead and the Cys491 sulfur atom is plotted for FGFR2. (D) A representative frame from the **6**-bound FGFR1 simulation after the extended P-loop conformation was induced. The ligand (salmon) and the P-loop are shown in sphere representation. (E) A representative frame from **6**-bound FGFR2 is shown.

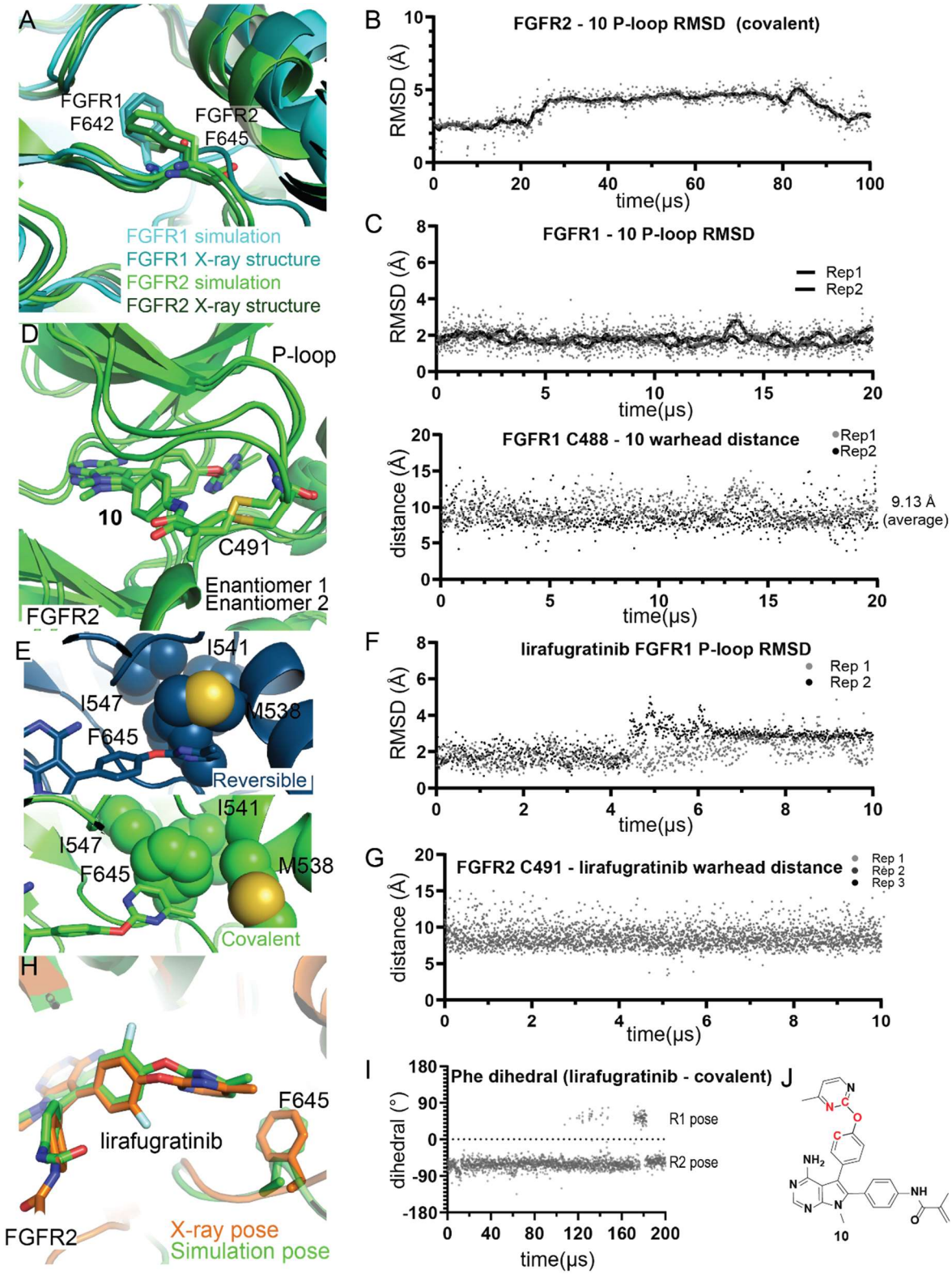


Figure S3: *Lirafugratinib induces similar protein conformational changes to those induced by*

*compound **10** upon covalent engagement.* (A) Representative frames from FGFR1 (cyan) and FGFR2 (green) apo simulations are superposed with apo X-ray structures of FGFR1 (PDB ID: 4UWY) and FGFR2 (PDB ID: 1GJO). (B) The P-loop RMSD (dots, with the solid line a guide to the eye) is plotted from a simulation of **10** covalently bound with FGFR2, using the same atom selections as Figure S1 for superposition and RMSD calculation; the X-ray structure of **10**-bound FGFR2 is used as the reference. (C) The P-loop RMSD is plotted from a simulation of **10** reversibly bound with FGFR1, using the same atom selections as in Figure S1 for superposition and RMSD calculation, and using the first frame of the simulation as the reference (top; for this and subsequent panels, the results from multiple independent simulations are shown). Distance between the **10** warhead and the FGFR1 Cys488 sulfur atom is plotted (bottom). (D) A representative frame is superposed from each of two simulations of **10** covalently bound with FGFR2, in which the covalent bond was formed using alternative enantiomers of the **10** warhead. (E) Representative frames from simulations of 10 reversibly (dark, top panel) and covalently (light, bottom panel) bound to FGFR2. Back pocket residues packing with Phe645 and **10** are shown in sphere representation. (F) The P-loop RMSD is plotted from a simulation of lirafugratinib reversibly bound with FGFR1, using the same atom selections as in Figure S1 for superposition and RMSD calculations, and using the first frame of the simulation as the reference. (G) FGFR2 Cys491 sulfur- lirafugratinib warhead distance is plotted from a simulation of lirafugratinib reversibly bound with FGFR2. (H) A representative pose from a simulation of lirafugratinib covalently bound with FGFR2 is superposed on the X-ray structure. (I) The F645 N-Cα-Cβ-Cγ dihedral is plotted for a simulation of lirafugratinib covalently bound with FGFR2. (J) The ligand dihedral angle presented in the plots in Figure 3 E and F is shown.

Supplemental Table

Simulation length	Box size	Number of atoms	Referenced figure	Referenced Movie	System description	Description
3 X 25 µs	~81 Å	~54 K	Figure 1A, B; Figure S1A, B; Figure 3A and Figure S3A	Movie 1	FGFR1 residues 460-764	FGFR1 Apo
3 X 25 µs	~78 Å	~47.5K	Figure 1A, B; Figure S1A, B; Figure 3A and Figure S3A	Movie 2	FGFR2 residues 467-764	FGFR2 Apo
3 X 10 µs	~84 Å	~60 K	Figure 1C, D and Figure S1C		FGFR1 residues 460-764	FGFR1 - Futibatinib
1 X 10 µs	~88 Å	~68 K	Figure 2 B, C, D and S2B, D	Movie 3	FGFR1 residues 460-764	FGFR1 - Compound 6
1 X 10 µs	~88 Å	~68 K	Figure S2B, C, E	Movie 4	FGFR2 residues 467-764	FGFR2 - Compound 6
1 X 10 µs	~88 Å	~68 K	Figure 2D	Movie 5	FGFR1 residues 460-764	FGFR1 - Compound 7
1 X 10 µs	~88 Å	~68 K	Figure 2D	Movie 6	FGFR1 residues 460-764	FGFR1 - Compound 8
2 X 10 µs	~84 Å	~60 K	Figure S3C		FGFR1 residues 460-764	FGFR1 - Compound 10
1 X 100 µs	~83 Å	~57 K	Figure 3B, E, H, I and Figure S3E	Movie 7	FGFR2 residues 467-764	FGFR2 - Compound 10 reversible
1 X 100 µs	~84 Å	~60 K	Figure 3C, F, I and Figure S3B, E	Movie 7	FGFR2 residues 467-764	FGFR2 - Compound 10 covalent
1 X 100 µs	~84 Å	~60 K	Figure S3D		FGFR2 residues 467-764	FGFR2 - Compound 10 covalent enantiomer 2
1 X 200 µs	~84 Å	~60 K	Figure 3D		FGFR2 residues 467-764	FGFR2 - Apo from Compound 10 covalent
2 X 10 µs	~84 Å	~60 K	Figure S3F		FGFR1 residues 460-764	FGFR1 - lirafugratinib
3 X 10 µs	~88 Å	~68 K	Figure S3G		FGFR2 residues 467-764	FGFR2 - lirafugratinib
1 X 200 µs	~84 Å	~60 K	Figure S3H, I		FGFR2 residues 467-764	FGFR2 - lirafugratinib

Table S1: *List of MD simulations*

Movie captions

Supplemental Movie 1

Simulation of the apo FGFR1 kinase domain. FGFR1 is colored in cyan. The view is focused on the P-loop, and the vicinity of Cys488 is highlighted in a darker shade of blue.

Supplemental Movie 2

Simulation of the apo FGFR2 kinase domain. FGFR2 is colored in green. The view is focused on the P-loop, and the vicinity of Cys 491 is highlighted in a darker shade of green.

Supplemental Movie 3

Simulation of the FGFR1 kinase domain bound to compound **6**, starting from a disordered P-loop conformation. Bound to compound **6**, the FGFR1 P-loop adapts an extended conformation. FGFR1 is colored in cyan.

Supplemental Movie 4

Simulation of the FGFR2 kinase domain bound to compound **6**. P-loop Cys491 can sample close warhead distances that likely allow targeting of Cys491. FGFR2 is colored in green.

Supplemental Movie 5

Simulation of the FGFR1 kinase domain bound to compound **7**, starting from an extended P-loop conformation. Bound to compound **7**, the FGFR1 P-loop becomes disordered. FGFR1 is colored in cyan.

Supplemental Movie 6

Simulation of the FGFR1 kinase domain bound to compound **8**, starting from an extended P-loop

conformation. Bound to compound **8**, the FGFR1 P-loop becomes disordered. FGFR1 is colored in cyan.

Supplemental Movie 7

Aggregate trajectory of two simulations: (i) Simulation of the FGFR2 kinase domain bound to compound **10**. In this simulation, we observed that FGFR2 P-loop Cys491 can sample close distances to the warhead methyl acrylamide C atom, likely allowing targeting of Cys491. (ii) A second simulation was started from a frame selected from the initial simulation, and the covalent bond between the warhead and the kinase was manually built. Superposition of a pose from the second simulation is shown with the X-ray structure pose of FGFR2 bound to **10**. FGFR2 is colored in green.

Chemistry

Chemical synthesis was conducted at Pharmaron Beijing Co., Ltd.

General Methods

Commercial Reagent/solvent sources

The known starting materials can be purchased from TCI, PharmaBlock, Enamine, and other commercial sources.

Set up of air sensitive reactions

The reactions were carried out under nitrogen atmosphere, a reaction vial or three necked bottle was charged with the materials and a stirbar before being evacuated and purged with nitrogen three times.

Instrumentation

Standard labware was used to perform chemical reactions, including oil bath, magnetic stir plate with heating, rotary evaporator, glass manifold, etc.

Methods for analytical and prep HPLC, LC-MS

1) Thin layer chromatography (TLC):

The developing solvent system used in the reactions included: A: DCM/MeOH, B: Petroleum ether/EtOAc. The ratio of the volume of the solvent was adjusted according to the polarity of the compounds, and a small quantity of alkaline reagent such as trimethylamine or acidic reagent such as acetic acid can also be added for adjustment.

2) Prep-HPLC:

Method A: Column: Sunfire prep C18 Column, 30*150mm, 5um; Mobile Phase A: Water (0.1% FA), Mobile Phase B: ACN; Flow rate: 60 mL/min mL/min; Wavelength: 254nm/220nm;

Method B: Column: XBridge Prep OBD C18 Column, 30*150 mm, 5μm; Mobile Phase A: Water (10mmol/L NH_4HCO_3) + 0.05% NH_3, Mobile Phase B: ACN; Flow rate: 60 mL/min mL/min; Wave Length: 254 nm/220 nm;

Method C: Column: Sunfire prep C18 Column, 30*150 mm, 5 μm; Mobile Phase A: Water (0.05% FA), Mobile Phase B: ACN; Flow rate: 60 mL/min mL/min; Wavelength: 254 nm/220 nm;

3) LCMS:

Method A: LCMS45/185/126

Typically, the analytical LC-MS system is equipped with Shimadzu LCMS-2020, PDA detector (operating at 254 nm), ELSD detector, and ESI-source operating in positive ion mode. LC-conditions: The column is HALO C18 30*3.0 mm, 2 μm operating at 40 °C with 1.5 mL/min of a binary gradient consisting of water + 0.1% Formic acid (A) and acetonitrile + 0.1% Formic acid (B）. The retention times (tR) are expressed in minutes based on UV-trace at 254 nm.

Gradient:

0.01 min 5% B

1.00 min 100% B

1.40 min 100% B

1.42 min 5% B

Total run time: 1.5 min

Method B: LCMS72/94/127/186

Typically, the analytical LC-MS system is equipped with Shimadzu LCMS-2020, PDA detector (operating at 254 nm), ELSD detector, and ESI-source operating in positive ion mode. LC-conditions: The column is Shim-pack Scepter C18-120,33*3.0 mm, 3 um particles operating at 30 °C with 1.5 mL/min of a binary gradient consisting of water + 5 mM NH_4HCO_3 (A) and acetonitrile (B). The retention times (tR) are expressed in minutes based on UV-trace at 254 nm.

Gradient:

0.01 min 10% B

1.20 min 95% B

1.80 min 95% B

1.82 min 10% B

Total run time: 2.0 min

Method C: LCMS68/17/19/82/136

Typically, the analytical LC-MS system is equipped with Shimadzu LCMS-2020, PDA detector (operating at 254 nm), ELSD detector, and ESI-source operating in positive ion mode. LC-conditions: The column is HALO C18 30*3.0mm, 2 μm operating at 40 °C with 1.5 mL/min of a binary gradient consisting of water + 0.05 % trifluoroacetic acid (A) and acetonitrile + 0.05 % trifluoroacetic acid (B). The retention times (tR) are expressed in minutes based on UV-trace at 254 nm.

Gradient:

0.01 min 5% B

1.20 min 100% B

1.80 min 100% B

1.82 min 5% B

Total run time: 2.0 min

Method D: LCMS137

Typically, the analytical LC-MS system is equipped with Shimadzu LCMS-2020, PDA detector (operating at 254 nm), ELSD detector, and ESI-source operating in positive ion mode. LC-conditions: The column is Shim-pack ScepterC18-120,33*3.0mm, 3um operating at 30 °C with 1.5 mL/min of a binary gradient consisting of water + 6.5 mM NH_4HCO_3 + Ammonia (pH = 10) (A) and acetonitrile (B). The retention times (tR) are expressed in minutes based on UV-trace at 254 nm.

Gradient:

0.01 min 10% B

1.20 min 95% B

1.80 min 95% B

1.82 min 10% B

Total run time: 2.0 min

NMR

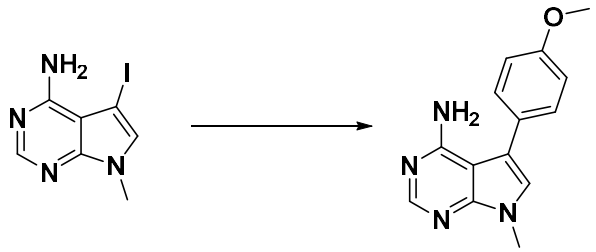
Model: AVANCE III HD 400 MHz and AVANCE NEO 400 MHz

Manufacture: Bruker

Chemical shifts are expressed in parts per million (ppm, δ units). Coupling constants are in units of hertz (Hz). Splitting patterns describe apparent multiplicities and are designated as s (singlet), d (doublet), t (triplet), q (quartet), quint (quintet), m (multiplet), br (broad).

Synthetic Procedures

5-(4-methoxyphenyl)-7-methyl-7H-pyrrolo[2,3-d]pyrimidin-4-amine (1)

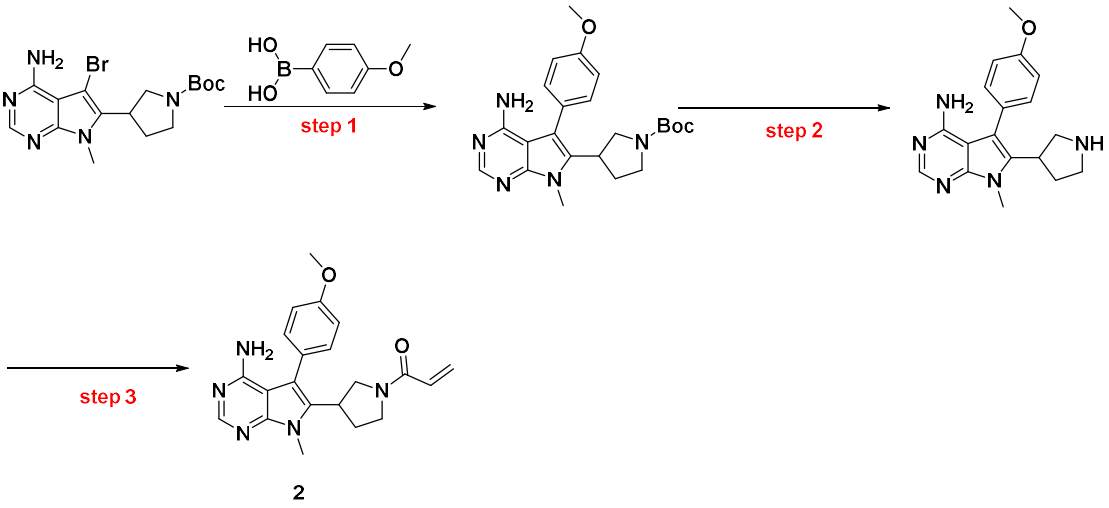


1

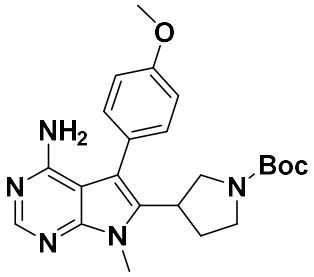
A resealable reaction vial was charged with 5-iodo-7-methyl-7H-pyrrolo[2,3-d]pyrimidin-4-amine (20 g, 72.9 mmol), [4-(methoxycarbonyl)phenyl]boronic acid (15.7 g, 87.4 mmol), $Pd(DtBPF)Cl_2$ (4.74 g, 7.29 mmol), CsF (33.1 g, 218 mmol), DMF (200 mL), H_2O (25 mL) and a stir bar before being evacuated and purged with nitrogen three times. The mixture was stirred for 1 h at 90 °C. The reaction mixture was diluted with H_2O (500 mL), and the aqueous phase was extracted with DCM (200 mL) three times. The combined organic layers were washed with

brines, dried over sodium sulfate, filtered, and concentrated in vacuo. The reaction mixture was added MeCN (10 mL) and filtered through a pad of Celite, the pad was washed with MeCN, the solid is 5-(4-methoxyphenyl)-7-methyl-7H-pyrrolo[2,3-d]pyrimidin-4-amine **1** (11.0 g, 53%) as a yellow amorphous solid. LC/MS(BAS1): $[M+H]^+$= 255.10; t_R = 0.897 min. ^{1}H NMR (400 MHz, DMSO-d_6) δ 8.14 (s, 1H), 7.37 (d, J = 8.3 Hz, 2H), 7.22 (s, 1H), 7.04 (d, J = 8.5 Hz, 2H), 3.80 (s, 3H), 3.73 (s, 3H).

1-(3-(4-amino-5-(4-methoxyphenyl)-7-methyl-7H-pyrrolo[2,3-d]pyrimidin-6-yl)pyrrolidin-1-yl)prop-2-en-1-one (2)

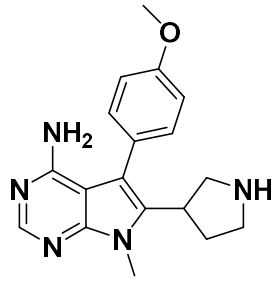


tert-butyl 3-(4-amino-5-(4-methoxyphenyl)-7-methyl-7H-pyrrolo[2,3-d]pyrimidin-6-yl)pyrrole dine-1-carboxylate



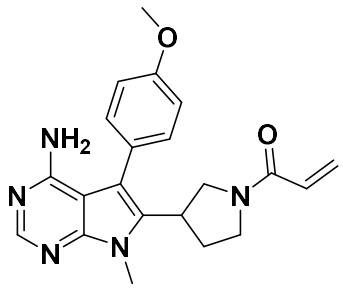
A resealable reaction vial was charged with tert-butyl 3-(4-amino-5-bromo-7-methyl-7H-pyrrolo[2,3-d]pyrimidin-6-yl)pyrrolidine-1-carboxylate (100 mg, 0.25 mmol), (4-methoxyphenyl)boronic acid (46 mg, 0.30 mmol), Pd(DtBPF)Cl_2 (16.3 mg, 0.025 mmol), CsF (115 mg, 0.757 mmol), DMF (2 mL), H_2O (0.25 mL) and a stir bar before being evacuated and purged with nitrogen three times. The mixture was stirred for 1 h at 90 °C. The reaction mixture was diluted with H_2O (10 mL), and the aqueous phase was extracted with DCM (10 mL) three times. The combined organic layers were washed with brines, dried over sodium sulfate, filtered, and concentrated in vacuo. The resulting crude material was purified by HPLC. Concentration in vacuo resulted in tert-butyl 3-(4-amino-5-(4-methoxyphenyl)-7-methyl-7H-pyrrolo[2,3-d]pyrimidin-6-yl)pyrrolidine-1-carboxylate (50 mg, 46.79%) as a yellow solid.

5-(4-methoxyphenyl)-7-methyl-6-(pyrrolidin-3-yl)-7H-pyrrolo[2,3-d]pyrimidin-4-amine



A round bottomed flask was charged with tert-butyl 3-(4-amino-5-(4-methoxyphenyl)-7-methyl-7H-pyrrolo[2,3-d]pyrimidin-6-yl)pyrrolidine-1-carboxylate (40 mg, 0.094 mmol), DCM (2 mL) and a stir bar. TFA (0.5 mL) was added. The reaction mixture was stirred for 1 h at room temperature. The solvent was removed in vacuo resulted in 5-(4-methoxyphenyl)-7-methyl-6-(pyrrolidin-3-yl)-7H-pyrrolo[2,3-d]pyrimidin-4-amine (30 mg, 100%) as a dark oil.

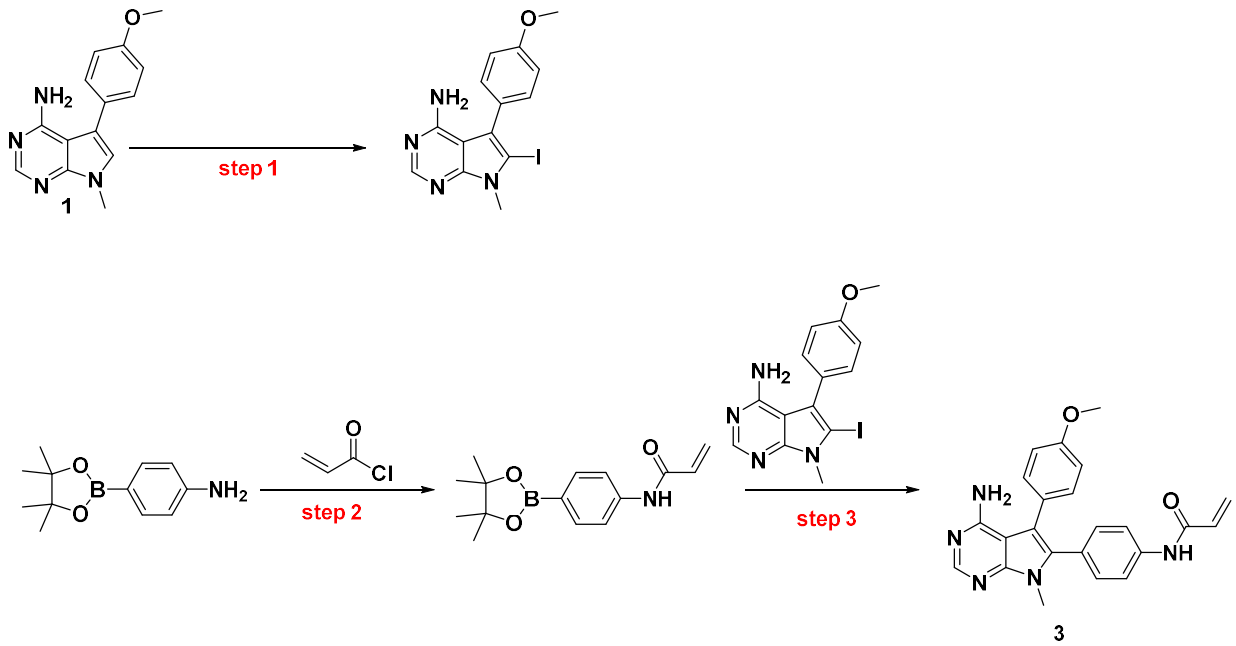
1-(3-(4-amino-5-(4-methoxyphenyl)-7-methyl-7H-pyrrolo[2,3-d]pyrimidin-6-yl)pyrrolidin-1-yl)prop-2-en-1-one (**2**)



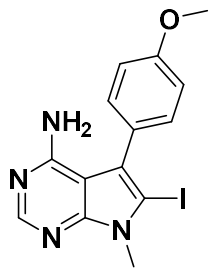
A round bottomed flask was charged with 5-(4-methoxyphenyl)-7-methyl-6-(pyrrolidin-3-yl)-7H-pyrrolo[2,3-d]pyrimidin-4-amine (30 mg, 0.093 mmol), Et_3N (28.2 mg, 0.28 mmol) DCM (2 mL) and a stir bar. The mixture was cooled to -15 °C, prop-2-enoyl chloride (9.24 mg, 0.1 mmol) was added dropwise and the solution was stirred for 1 h at -15 °C. The reaction mixture was quenched with MeOH, concentrated in vacuo. The resulting crude material was purified by Prep-HPLC. (Column: XBridge Shield RP18 OBD Column, 30*150 mm, 5μm; Mobile Phase A: Water (10 mmol/L NH_4HCO_3), Mobile Phase B: ACN; Flow rate: 60 mL/min; Gradient: 15% B to 40% B in 7 min, 40% B; Wavelength: 254 nm; RT1(min): 6.2; Number Of Runs: 0) Lyophilization yielded 1-(3-(4-amino-5-(4-methoxyphenyl)-7-methyl-7H-pyrrolo[2,3-

d]pyrimidin-6-yl)pyrrolidin-1-yl)prop-2-en-1-one **2** (20 mg, 57.1 %) as a white amorphous solid. LC/MS(BAS1): $[M+H]^+$= 378.15; t_R =1.07 min. ^{1}H NMR (400 MHz, DMSO-d_6) δ 8.10 (d, J = 1.7 Hz, 1H), 7.30 (d, *J* = 8.2 Hz, 2H), 7.03 (d, *J* = 8.3 Hz, 2H), 6.47 (ddd, *J* = 40.7, 16.8, 10.3 Hz, 1H), 6.09 (ddd, *J* = 16.8, 5.6, 2.4 Hz, 1H), 5.63 (ddd, *J* = 14.6, 10.3, 2.5 Hz, 2H), 3.81 (d, *J* = 1.1 Hz, 3H), 3.77 (d, *J* = 6.0 Hz, 4H), 3.66 (q, *J* = 8.7, 7.2 Hz, 1H), 3.58 – 3.37 (m, 2H), 2.25 – 1.88 (m, 2H).

N-(4-(4-amino-5-(4-methoxyphenyl)-7-methyl-7H-pyrrolo[2,3-d]pyrimidin-6-yl)phenyl)acrylamide (3)

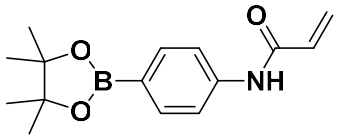


6-iodo-5-(4-methoxyphenyl)-7-methyl-7H-pyrrolo[2,3-d]pyrimidin-4-amine



A round bottomed flask was charged with 5-(4-methoxyphenyl)-7-methyl-7H-pyrrolo[2,3-d]pyrimidin-4-amine (1 g, 3.93 mmol), DCM (20 mL) and TFA (0.5 mL) and a stir bar. 1-iodopyrrolidine-2,5-dione (1.06 g, 4.71 mmol) was added, and the solution was stirred for 1 h at room temperature. The reaction mixture was diluted with Na_2SO_3 solution (100 mL), and the aqueous phase was extracted with DCM (100 mL) three times. The combined organic layers were washed with saturated brines, dried over sodium sulfate, filtered, and concentrated in vacuo. The resulting crude material was purified by HPLC. Concentration in vacuo resulted in 6-iodo-5-(4-methoxyphenyl)-7-methyl-7H-pyrrolo[2,3-d]pyrimidin-4-amine (1 g, 66.8 %) as a yellow solid.

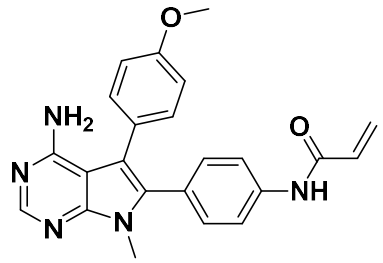
N-(4-(4,4,5,5-tetramethyl-1,3,2-dioxaborolan-2-yl) phenyl) acrylamide



A round bottomed flask was charged with 4-(4,4,5,5-tetramethyl-1,3,2-dioxaborolan-2-yl)aniline (5 g, 22.82 mmol), prop-2-enoyl chloride (2.07 g, 22.82 mmol), TEA (6.93 g, 68.46 mmol), dichloromethane (100 mL) and a stirbar. The solution was stirred for 1 h at 0 °C. The reaction

mixture was quenched with water, extracted with DCM. The organic phase was dried over Na_2SO_4, filtered and evaporated in vacuo. The resulting crude material was purified by silica gel chromatography. Concentration in vacuo resulted in N-(4-(4,4,5,5-tetramethyl-1,3,2-dioxaborolan-2-yl)phenyl)acrylamide (5 g, 80.21%) as a yellow oil.

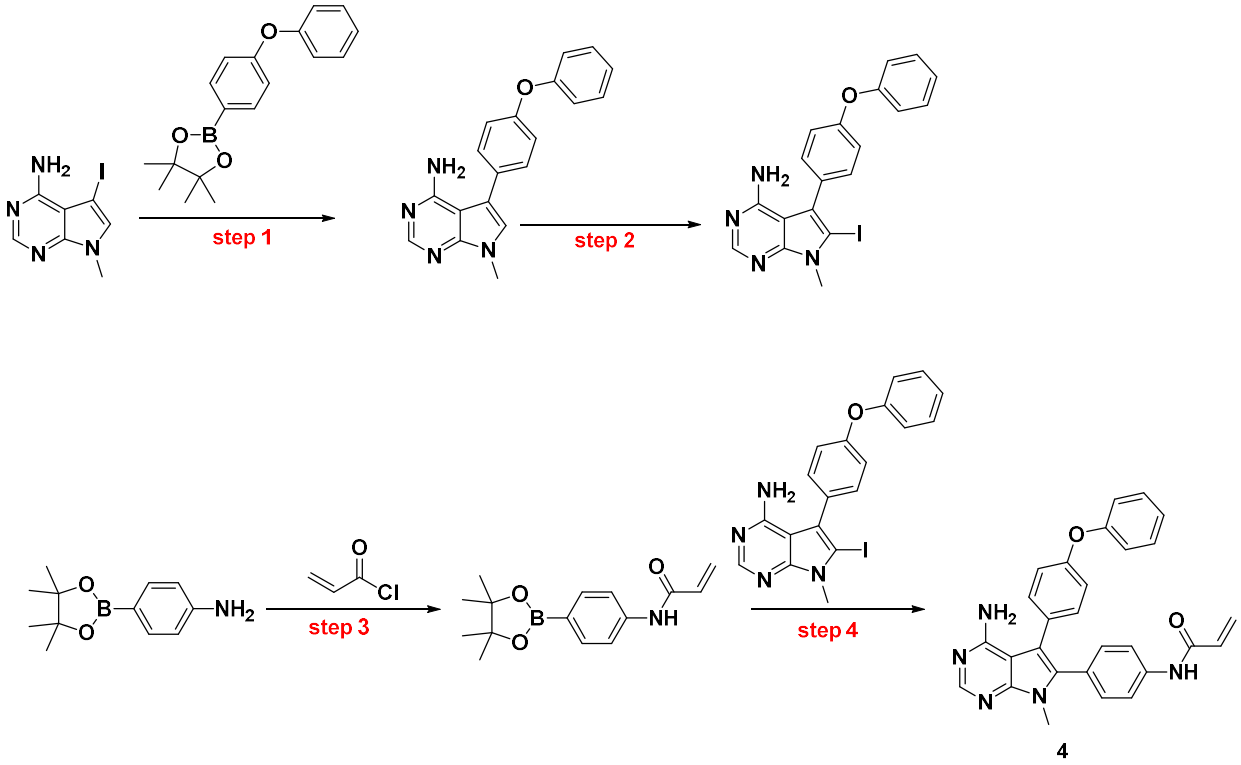
N-(4-(4-amino-5-(4-methoxyphenyl)-7-methyl-7H-pyrrolo[2,3-d]pyrimidin-6-yl)phenyl)acrylamide (**3**)



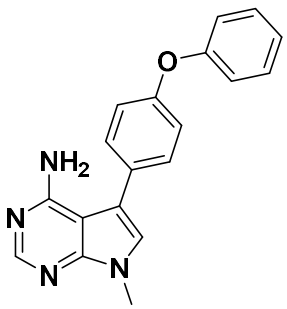
A resealable reaction vial was charged with 6-iodo-5-(4-methoxyphenyl)-7-methyl-7H-pyrrolo[2,3-d]pyrimidin-4-amine (100 mg, 0.26 mmol), N-(4-(4,4,5,5-tetramethyl-1,3,2-dioxaborolan-2-yl)phenyl)acrylamide (86.2 mg, 0.32 mmol), Pd(DtBPF)Cl_2 (17 mg, 0.026 mmol), CsF (119.8 mg, 0.79 mmol), DMF (2 mL), H_2O (0.25 mL) and a stir bar before being evacuated and purged with nitrogen three times. The mixture was stirred for 1 h at 90 °C. The reaction mixture was diluted with H_2O (10 mL), and the aqueous phase was extracted with DCM (10 mL) three times. The combined organic layers were washed with brines, dried over sodium sulfate, filtered, and concentrated in vacuo. The resulting crude material was purified by Prep-HPLC. (Column: XBridge Shield RP18 OBD Column, 30*150 mm, 5 μm; Mobile Phase A: Water (10 mmol/L NH_4HCO_3), Mobile Phase B: ACN; Flow rate: 60 mL/min; Gradient: 25% B to 45% B in 7 min, 45% B; Wavelength: 254/220 nm; RT1(min): 6.5). Lyophilization yielded N-(4-(4-amino-5-(4-methoxyphenyl)-7-methyl-7H-pyrrolo[2,3-d]pyrimidin-6-yl)phenyl)acrylamide **3** (26.3 mg, 25 %) as a white amorphous solid. LC/MS(BAS1):$[M+H]^+$= 400.20; t_R = 1.394 min.

^{1}H NMR (400 MHz, DMSO-d_6) δ 10.26 (s, 1H), 8.18 (s, 1H), 7.70 – 7.64 (m, 2H), 7.32 – 7.25 (m, 2H), 7.21 – 7.12 (m, 2H), 6.96 – 6.90 (m, 2H), 6.44 (dd, J = 17.0, 10.1 Hz, 1H), 6.27 (dd, J = 16.9, 2.1 Hz, 1H), 5.84 (s, 1H), 5.78 (dd, J = 10.0, 2.1 Hz, 1H), 3.75 (s, 3H), 3.60 (s, 3H).

N-(4-(4-amino-7-methyl-5-(4-phenoxyphenyl)-7H-pyrrolo[2,3-d]pyrimidin-6-yl)phenyl)acrylamide (4)

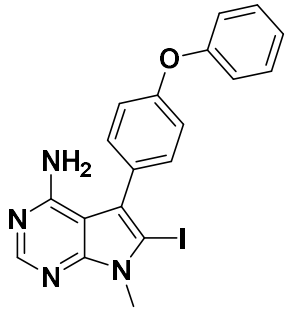


7-methyl-5-(4-phenoxyphenyl)-7H-pyrrolo[2,3-d]pyrimidin-4-amine



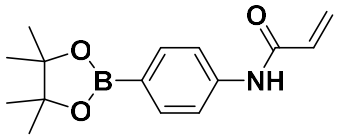
A resealable reaction vial was charged with 5-iodo-7-methyl-7H-pyrrolo[2,3-d]pyrimidin-4-amine (200 mg, 0.73 mmol), 4,4,5,5-tetramethyl-2-(4-phenoxyphenyl)-1,3,2-dioxaborolane (259.2 mg, 0.876 mmol), $Pd(dppf)Cl_2$ (53.3 mg, 0.073 mmol), K_3PO_4 (464.2 mg, 2.19 mmol), DMF (4 mL), H_2O (0.5 mL) and a stir bar before being evacuated and purged with nitrogen three times. The mixture was stirred for 1 h at 90 °C. The reaction mixture was diluted with H_2O (10 mL), and the aqueous phase was extracted with EA (10 mL) three times. The combined organic layers were washed with brines, dried over sodium sulfate, filtered, and concentrated in vacuo. The resulting crude material was purified by HPLC. Concentration in vacuo resulted in 7-methyl-5-(4-phenoxyphenyl)-7H-pyrrolo[2,3-d]pyrimidin-4-amine (120.0 mg, 52.0%) as a yellow solid.

6-iodo-7-methyl-5-(4-phenoxyphenyl)-7H-pyrrolo[2,3-d]pyrimidin-4-amine



A round bottomed flask was charged with 7-methyl-5-(4-phenoxyphenyl)-7H-pyrrolo[2,3-d]pyrimidin-4-amine (120.0 mg, 0.379 mmol), DCM (4 mL) and TFA (0.2 mL) and a stirbar. 1-iodopyrrolidine-2,5-dione (102.5 mg, 0.455 mmol) was added, and the solution was stirred for 1 h at room temperature. The reaction mixture was diluted with Na_2SO_3 solution (10 mL), and the aqueous phase was extracted with DCM (10 mL) three times. The combined organic layers were washed with saturated brines, dried over sodium sulfate, filtered, and concentrated in vacuo. The resulting crude material was purified by HPLC. Concentration in vacuo resulted in 6-iodo-7-methyl-5-(4-phenoxyphenyl)-7H-pyrrolo[2,3-d]pyrimidin-4-amine (100 mg, 59.6 %) as a yellow solid.

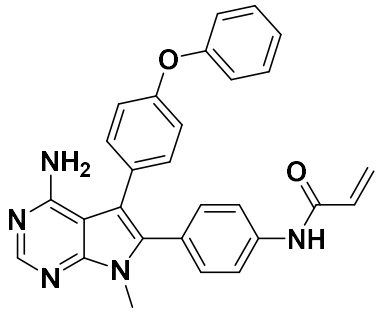
N-(4-(4,4,5,5-tetramethyl-1,3,2-dioxaborolan-2-yl)phenyl)acrylamide



A round bottomed flask was charged with 4-(4,4,5,5-tetramethyl-1,3,2-dioxaborolan-2-yl)aniline (1 g, 4.56 mmol), prop-2-enoyl chloride (413 mg, 4.56 mmol), TEA (1.39 g, 13.69 mmol), dichloromethane (20 mL) and a stirbar. The solution was stirred for 1 h at 0 °C. The reaction mixture was quenched with water, extracted with DCM. The organic phase was dried over Na_2SO_4, filtered and evaporated in vacuo. The resulting crude material was purified by silica gel

chromatography. Concentration in vacuo resulted in N-(4-(4,4,5,5-tetramethyl-1,3,2-dioxaborolan-2-yl)phenyl)acrylamide (750 mg, 60.1%) as a yellow oil.

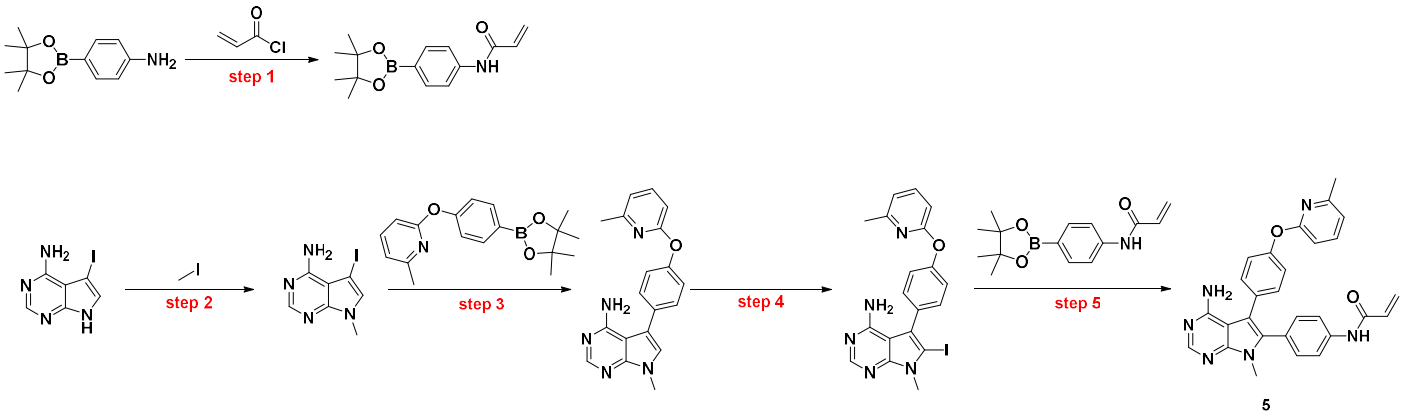
N-(4-(4-amino-7-methyl-5-(4-phenoxyphenyl)-7H-pyrrolo[2,3-d]pyrimidin-6-yl)phenyl)acrylamide (**4**)



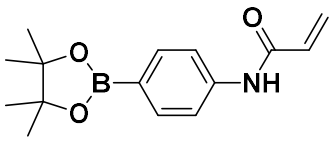
A resealable reaction vial was charged with 6-iodo-7-methyl-5-(4-phenoxyphenyl)-7H-pyrrolo[2,3-d]pyrimidin-4-amine (100 mg, 0.23 mmol), N-(4-(4,4,5,5-tetramethyl-1,3,2-dioxaborolan-2-yl)phenyl)acrylamide (74.1 mg, 0.27 mmol), $Pd(DtBPF)Cl_2$ (13 mg, 0.02 mmol), CsF (103 mg, 0.69 mmol), DMF (2 mL), H_2O (0.25 mL) and a stir bar before being evacuated and purged with nitrogen three times. The mixture was stirred for 1 h at 90 °C. The reaction mixture was diluted with H_2O (10 mL), and the aqueous phase was extracted with DCM (10 mL) three times. The combined organic layers were washed with brines, dried over sodium sulfate, filtered, and concentrated in vacuo. The resulting crude material was purified by Prep-HPLC. (Column: XBridge Shield RP18 OBD Column, 30*150 mm, 5μm; Mobile Phase A: Water(10 mmol/L NH_4HCO_3), Mobile Phase B: ACN; Flow rate: 60 mL/min; Gradient: 35% B to 48% B in 7 min, 48% B; Wave Length: 254/220 nm; RT1(min): 6.5). Lyophilization yielded N-(4-(4-

amino-7-methyl-5-(4-phenoxyphenyl)-7H-pyrrolo[2,3-d]pyrimidin-6-yl)phenyl)acrylamide **4** (27.9 mg, 26.7 %) as a white amorphous solid. LC/MS(BAS1): $[M+H]^+$= 462.15; tR =1.609 min. ^{1}H NMR (400 MHz, DMSO-d_6) δ 10.28 (s, 1H), 8.19 (s, 1H), 7.69 (d, J = 8.6 Hz, 2H), 7.45 – 7.36 (m, 2H), 7.29 (d, J = 8.7 Hz, 2H), 7.28 – 7.20 (m, 2H), 7.20 – 7.11 (m, 1H), 7.10 – 7.04 (m, 2H), 7.01 – 6.94 (m, 2H), 6.45 (dd, J = 17.0, 10.1 Hz, 1H), 6.28 (dd, J = 17.0, 2.1 Hz, 1H), 5.84 (s, 1H), 5.78 (dd, J = 10.0, 2.1 Hz, 1H), 3.61 (s, 3H).

N-(4-(4-amino-7-methyl-5-(4-((6-methylpyridin-2-yl)oxy)phenyl)-7H-pyrrolo[2,3-d]pyrimidin-6-yl)phenyl)acrylamide (5)

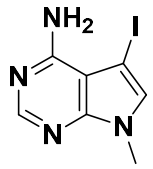


N-(4-(4,4,5,5-tetramethyl-1,3,2-dioxaborolan-2-yl) phenyl) acrylamide



A round bottomed flask was charged with 4-(4,4,5,5-tetramethyl-1,3,2-dioxaborolan-2-yl)aniline (1 g, 4.56 mmol), prop-2-enoyl chloride (413 mg, 4.56 mmol), triethyl amine (1.39 g, 13.69 mmol), dichloromethane (20 mL) and a stirbar. The solution was stirred for 1 h at 0 °C. The reaction mixture was quenched with water, extracted with DCM. The organic phase was dried over Na_2SO_4, filtered and evaporated in vacuo. The resulting crude material was purified by silica gel chromatography. Concentration in vacuo resulted in N-(4-(4,4,5,5-tetramethyl-1,3,2-dioxaborolan-2-yl)phenyl)acrylamide (750 mg, 60.1%) as a yellow oil.

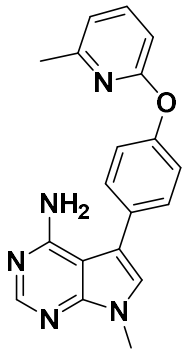
5-iodo-7-methyl-7H-pyrrolo[2,3-d]pyrimidin-4-amine



A round bottomed flask was charged with 5-iodo-7H-pyrrolo[2,3-d]pyrimidin-4-amine (50 g, 192.28 mmol), iodomethane (27.29 g, 192.28 mmol), Cs_2CO_3 (187.94 g, 576.84 mmol) and a stirbar. DMF (200 mL) was added, and the solution was stirred for 3 h at room temperature. The reaction mixture was diluted with H_2O (600 mL), and the aqueous phase was extracted with ethyl acetate (500 mL) three times. The combined organic layers were washed with saturated brines, dried over sodium sulfate, filtered, and concentrated in vacuo. The resulting crude material was purified by silica gel chromatography (eluting with PE/EA=1/2). Concentration in vacuo resulted

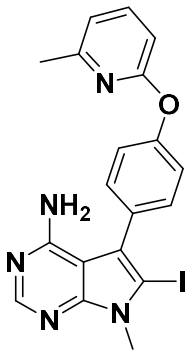
in 5-iodo-7-methyl-7H-pyrrolo[2,3-d]pyrimidin-4-amine (35 g, 66.4 %) as an off-white amorphous solid.

7-methyl-5-(4-((6-methylpyridin-2-yl)oxy)phenyl)-7H-pyrrolo[2,3-d]pyrimidin-4-amine



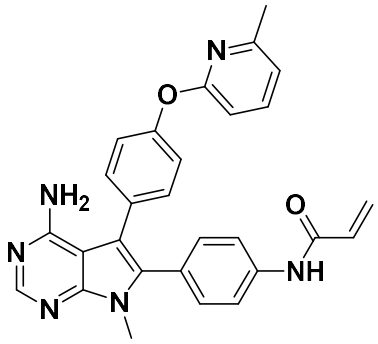
A resealable reaction vial was charged with 5-iodo-7-methyl-7H-pyrrolo[2,3-d]pyrimidin-4-amine (30 g, 109.46 mmol), 2-methyl-6-(4-(4,4,5,5-tetramethyl-1,3,2-dioxaborolan-2-yl)phenoxy)pyridine (40.88 g, 131.36 mmol), Pd(dppf)Cl2 (8 g, 10.95 mmol), K_3PO_4 (69.71 g, 328.39 mmol), DMF (300 mL), H_2O (37.5 mL) and a stir bar before being evacuated and purged with nitrogen three times. The mixture was stirred for 1 h at 90 °C. The reaction mixture was diluted with H_2O (900 mL), and the aqueous phase was extracted with EA (600 mL) three times. The combined organic layers were washed with brines, dried over sodium sulfate, filtered, and concentrated in vacuo. The reaction mixture was added MeCN (10 mL) and filtered through a pad of Celite, the pad was washed with MeCN, the solid is 7-methyl-5-(4-((6-methylpyridin-2-yl)oxy)phenyl)-7H-pyrrolo[2,3-d]pyrimidin-4-amine (22.0 g, 60.65%) as a yellow amorphous solid.

6-iodo-7-methyl-5-(4-((6-methylpyridin-2-yl)oxy)phenyl)-7H-pyrrolo[2,3-d]pyrimidin-4-amine



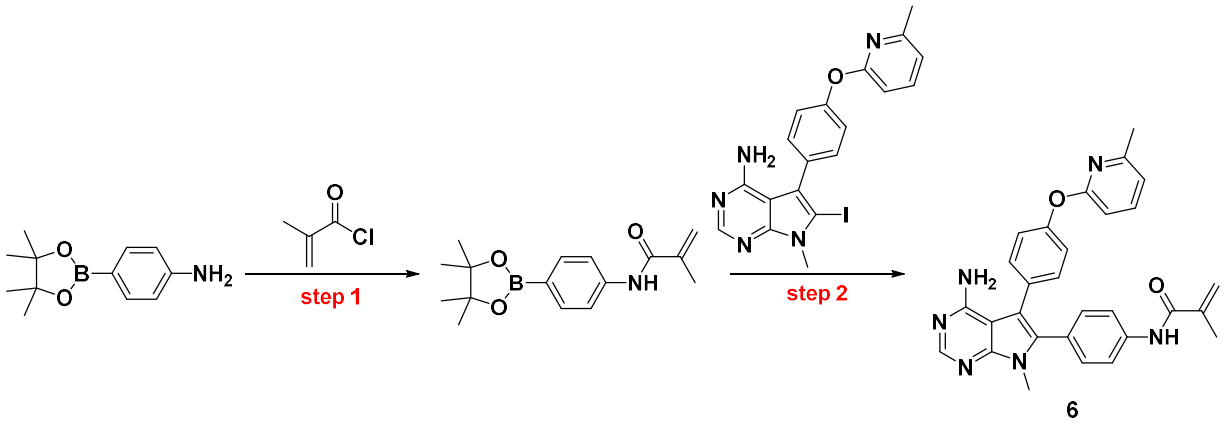
A round bottomed flask was charged with 7-methyl-5-(4-((6-methylpyridin-2-yl)oxy)phenyl)-7H-pyrrolo[2,3-d]pyrimidin-4-amine (20 g, 60.35 mmol), DCM (400 mL), TFA (20.65 g, 181.06 mmol) and a stir bar. The mixture was cooled to 0 °C, NIS (18.13 g, 66.39 mmol) was added, and the solution was stirred for 1 h at room temperature. The reaction mixture was diluted with saturated Na_2SO_3 solution, and the aqueous phase was extracted with DCM (400 mL) three times. The combined organic layers were washed with brines, dried over sodium sulfate, filtered, and concentrated in vacuo. DCM (20 mL) was added and the reaction mixture was filtered through a pad of Celite, the pad was washed with little DCM, the solid is 6-iodo-7-methyl-5-(4-((6-methylpyridin-2-yl)oxy)phenyl)-7H-pyrrolo[2,3-d]pyrimidin-4-amine (20 g, 72.47%) as an off-white amorphous solid.

N-(4-(4-amino-7-methyl-5-(4-((6-methylpyridin-2-yl)oxy)phenyl)-7H-pyrrolo[2,3-d]pyrimidin-6-yl)phenyl)acrylamide (**5**)

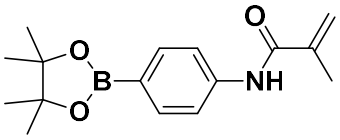


A resealable reaction vial was charged with 6-iodo-7-methyl-5-(4-((6-methylpyridin-2-yl)oxy)phenyl)-7H-pyrrolo[2,3-d]pyrimidin-4-amine (80 mg, 0.17 mmol), N-(4-(4,4,5,5-tetramethyl-1,3,2-dioxaborolan-2-yl)phenyl)acrylamide (57.3 mg, 0.21 mmol), Pd(DtBPF)Cl_2 (13 mg, 0.02 mmol), CsF (79.7 mg, 0.52 mmol), DMF (2 mL), H_2O (0.25 mL) and a stir bar before being evacuated and purged with nitrogen three times. The mixture was stirred for 1 h at 90 °C. The reaction mixture was diluted with H_2O (10 mL), and the aqueous phase was extracted with DCM (10 mL) three times. The combined organic layers were washed with brines, dried over sodium sulfate, filtered, and concentrated in vacuo. The resulting crude material was purified by Prep-HPLC. (Column: XBridge Prep Phenyl OBD Column, 19*250 mm, 5 μm; Flow rate: 25 mL/min; Gradient: 30% B to 60% B in 8 min, 60% B; Wavelength: 220 nm; RT1(min): 7.93. Lyophilization yielded N-(4-(4-amino-7-methyl-5-(4-((6-methylpyridin-2-yl)oxy)phenyl)-7H-pyrrolo[2,3-d]pyrimidin-6-yl)phenyl)acrylamide **5** (20 mg, 24 %) as a white amorphous solid. LC/MS(BAS1):$[M+H]^+$= 477.25; t_R =1.074 min. ^{1}H NMR (400 MHz, DMSO-d_6) δ 10.28 (s, 1H), 8.20 (s, 1H), 7.72 (dd, *J* = 15.6, 8.0 Hz, 3H), 7.35 – 7.27 (m, 2H), 7.26 (dq, *J* = 8.5, 2.4, 1.8 Hz, 2H), 7.13 – 7.05 (m, 2H), 7.01 (d, *J* = 7.3 Hz, 1H), 6.78 (d, *J* = 8.1 Hz, 1H), 6.44 (dd, *J* = 17.0, 10.1 Hz, 1H), 6.28 (dd, *J* = 17.0, 2.1 Hz, 1H), 5.93 (s, 1H), 5.78 (dd, *J* = 10.1, 2.1 Hz, 1H), 3.62 (s, 3H), 2.34 (s, 3H).

N-(4-(4-amino-7-methyl-5-(4-((6-methylpyridin-2-yl)oxy)phenyl)-7H-pyrrolo[2,3-d]pyrimidin-6-yl)phenyl)methacrylamide 6

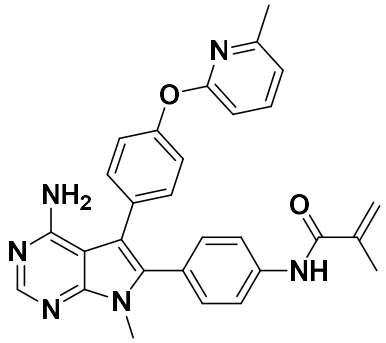


N-(4-(4,4,5,5-tetramethyl-1,3,2-dioxaborolan-2-yl)phenyl)methacrylamide



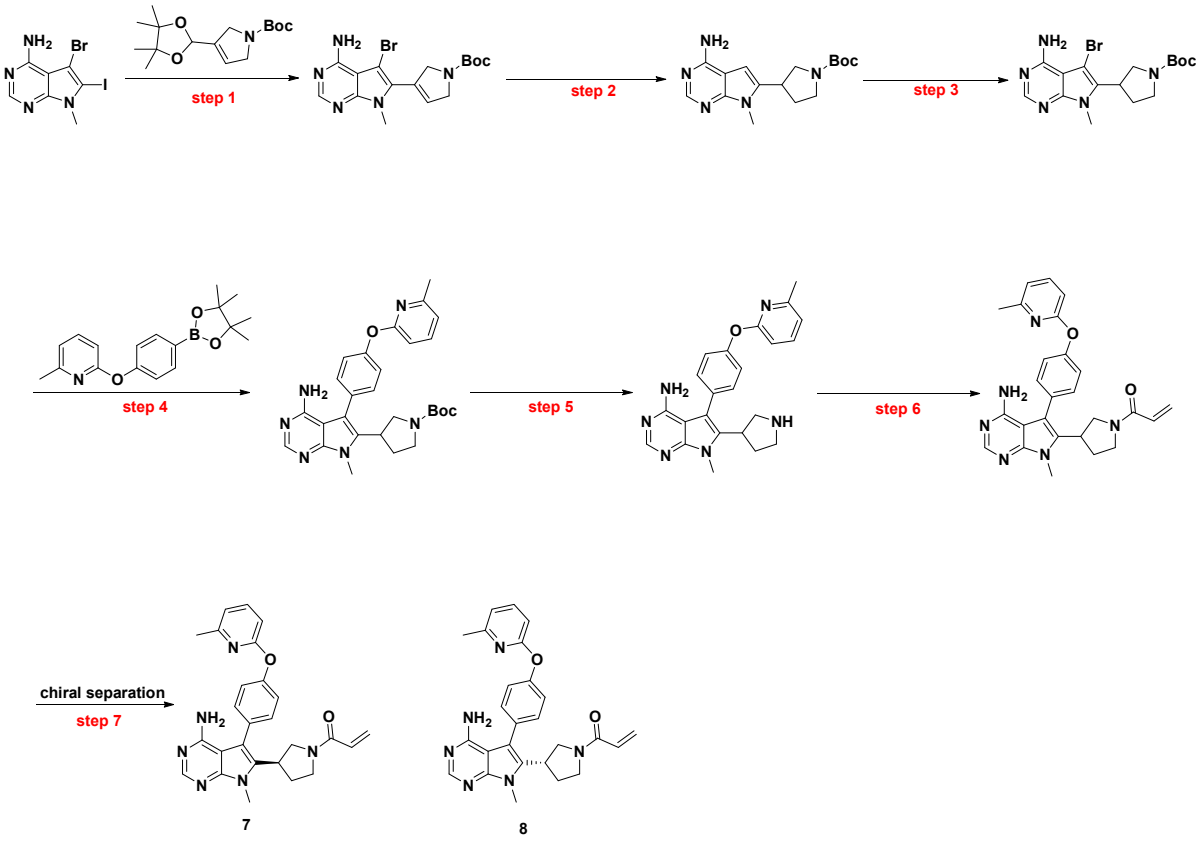
A round bottomed flask was charged with 4-(4,4,5,5-tetramethyl-1,3,2-dioxaborolan-2-yl)aniline (2.5 g, 11.4 mmol), DCM (50 mL), triethyl amine (3.46 g, 34.2 mmol) and a stirbar. The solution was cooled to 0 °C, methacryloyl chloride (1.31 g, 12.5 mmol) was added dropwise at 0 °C. After addition, the reaction mixture was stirred for 1 h at 0 °C. The reaction was quenched with water, extracted with DCM, dried over Na_2SO_4, evaporated in vacuo, the residue was purified by silica gel column chromatography, eluted with PE/EA (20:1 ~ 8:1) to afford N-(4-(4,4,5,5-tetramethyl-1,3,2-dioxaborolan-2-yl)phenyl)methacrylamide (2.9 g, 88.5%) as a white solid.

N-(4-(4-amino-7-methyl-5-(4-((6-methylpyridin-2-yl)oxy)phenyl)-7H-pyrrolo[2,3-d]pyrimidin-6-yl)phenyl)methacrylamide **6**

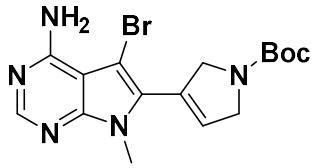


A resealable reaction vial was charged with 6-iodo-7-methyl-5-(4-((6-methylpyridin-2-yl)oxy)phenyl)-7H-pyrrolo[2,3-d]pyrimidin-4-amine (100 mg, 0.22 mmol), N-(4-(4,4,5,5-tetramethyl-1,3,2-dioxaborolan-2-yl)phenyl)methacrylamide (74.1 mg, 0.26 mmol), Pd(dppf)Cl_2 (16 mg, 0.02 mmol), K_3PO_4 (139.3 mg, 0.66 mmol), DMF (2 mL), H_2O (0.25 mL) and a stir bar before being evacuated and purged with nitrogen three times. The mixture was stirred for 1 h at 90 °C. The reaction mixture was diluted with H_2O (10 mL), and the aqueous phase was extracted with DCM (10 mL) three times. The combined organic layers were washed with brines, dried over sodium sulfate, filtered, and concentrated in vacuo. The resulting crude material was purified by Prep-HPLC. (Column: XBridge Prep OBD C18 Column, 30*150 mm, 5 μm; Flow rate: 60 mL/min; Gradient: 45% B to 70% B in 8 min, 70% B; Wave Length: 220 nm; RT1(min): 7.23). Lyophilization yielded N-(4-(4-amino-7-methyl-5-(4-((6-methylpyridin-2-yl)oxy)phenyl)-7H-pyrrolo[2,3-d]pyrimidin-6-yl)phenyl)methacrylamide **6** (15 mg, 13.9%) as a white amorphous solid. LC/MS(BAS1): $[M+H]^+$= 491.20; t_R =1.235 min. ^{1}H NMR (400 MHz, DMSO-d_6) δ 9.90 (s, 1H), 8.20 (s, 1H), 7.73 (t, *J* = 8.1 Hz, 3H), 7.33 – 7.22 (m, 4H), 7.13 – 7.05 (m, 2H), 7.01 (d, *J* = 7.3 Hz, 1H), 6.78 (d, *J* = 8.2 Hz, 1H), 5.91 (s, 1H), 5.80 (t, *J* = 1.0 Hz, 1H), 5.53 (t, *J* = 1.4 Hz, 1H), 3.62 (s, 3H), 2.35 (s, 3H), 1.95 (t, *J* = 1.2 Hz, 3H).

(R)-1-(3-(4-amino-7-methyl-5-(4-((6-methylpyridin-2-yl)oxy)phenyl)-7H-pyrrolo[2,3-d]pyrimidin-6-yl)pyrrolidin-1-yl)prop-2-en-1-one (7) and (S)-1-(3-(4-amino-7-methyl-5-(4-((6-methylpyridin-2-yl)oxy)phenyl)-7H-pyrrolo[2,3-d]pyrimidin-6-yl)pyrrolidin-1-yl)prop-2-en-1-one (8)

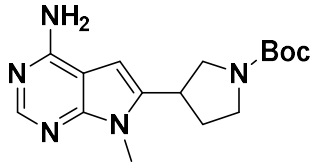


tert-butyl 3-(4-amino-5-bromo-7-methyl-7H-pyrrolo[2,3-d]pyrimidin-6-yl)-2,5-dihydro-1H-pyrrole-1-carboxylate



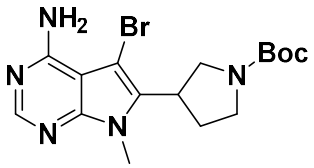
A resealable reaction vial was charged with 5-bromo-6-iodo-7-methyl-7H-pyrrolo[2,3-d]pyrimidin-4-amine (9 g, 25.5 mmol), tert-butyl 3-(4,4,5,5-tetramethyl-1,3-dioxolan-2-yl)-2,5-dihydro-1H-pyrrole-1-carboxylate (9.1 g, 30.6 mmol), $Pd(dppf)Cl_2$ (1.86 g, 2.55 mmol), K_3PO_4 (16.24 g, 76.5 mmol), DMF (160 mL), H_2O (20 mL) and a stir bar before being evacuated and purged with nitrogen three times. The mixture was stirred for 1 h at 90 °C. The reaction mixture was diluted with H_2O (600 mL), and the aqueous phase was extracted with EA (500 mL) three times. The combined organic layers were washed with brines, dried over sodium sulfate, filtered, and concentrated in vacuo. The resulting crude material was purified by silica gel chromatography (eluting with MeOH/DCM=1/30). Concentration in vacuo resulted in tert-butyl 3-(4-amino-5-bromo-7-methyl-7H-pyrrolo[2,3-d]pyrimidin-6-yl)-2,5-dihydro-1H-pyrrole-1-carboxylate (7 g, 69.6%) as a yellow solid.

tert-butyl 3-(4-amino-7-methyl-7H-pyrrolo[2,3-d]pyrimidin-6-yl)pyrrolidine-1-carboxylate



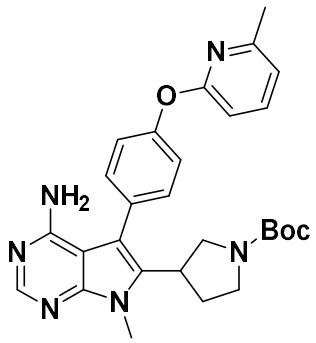
A resealable reaction vial was charged with tert-butyl 3-(4-amino-5-bromo-7-methyl-7H-pyrrolo[2,3-d]pyrimidin-6-yl)-2,5-dihydro-1H-pyrrole-1-carboxylate (6 g, 15.22 mmol), Pd-C (600 mg, 6 mol) and a stir bar before being evacuated and purged with hydrogen three times. MeOH (100 mL) was added, and the mixture was stirred overnight at 50 °C. The reaction mixture was filtered through a pad of Celite, the pad was washed with water, and the filtrate was concentrated in vacuo resulted in tert-butyl 3-(4-amino-7-methyl-7H-pyrrolo[2,3-d]pyrimidin-6-yl)pyrrolidine-1-carboxylate (4 g, 82.8%) as an off-white solid.

tert-butyl-3-(4-amino-5-bromo-7-methyl-7H-pyrrolo[2,3-d]pyrimidin-6-yl)pyrrolidine-1 carboxylate



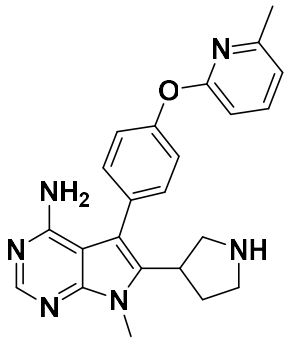
A round bottomed flask was charged with tert-butyl 3-(4-amino-7-methyl-7H-pyrrolo[2,3-d]pyrimidin-6-yl)pyrrolidine-1-carboxylate (3.8 g, 11.97 mmol), NBS (2.71 g, 11.97 mmol), and a stirbar. dichloromethane (50 mL) was added, and the solution was stirred 30 min at r.t. The reaction mixture was diluted with water (100 mL), and the aqueous phase was extracted with dichloromethane (150 mL) three times. The combined organic layers were washed with brine, dried over sodium sulfate, filtered, and concentrated in vacuo resulted in tert-butyl 3-(4-amino-5-bromo-7-methyl-7H-pyrrolo[2,3-d]pyrimidin-6-yl)pyrrolidine-1-carboxylate (3 g, 63.2%) as a light yellow solid.

tert-butyl-3-(4-amino-7-methyl-5-(4-((6-methylpyridin-2-yl)oxy)phenyl)-7H-pyrrolo[2,3-d]pyrimidin-6-yl)pyrrolidine-1-carboxylate



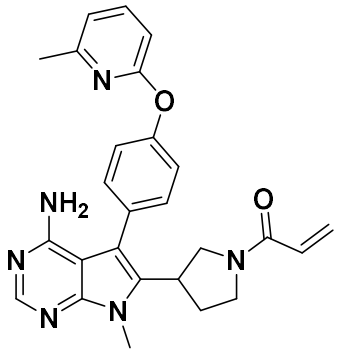
A resealable reaction vial was charged with in tert-butyl 3-(4-amino-5-bromo-7-methyl-7H-pyrrolo[2,3-d]pyrimidin-6-yl)pyrrolidine-1-carboxylate (1.5 g, 3.97 mmol), 2-methyl-6-(4-(4,4,5,5-tetramethyl-1,3,2-dioxaborolan-2-yl)phenoxy)pyridine (1.41 g, 4.54 mmol), $Pd(dtbpf)Cl_2$ (258.4 mg, 0.397 mmol), CsF (1.72 g, 11.36 mmol), DMF (20 mL), H_2O (2.5 mL) and a stirbar before being evacuated and purged with nitrogen three times. The mixture was stirred 2 h at 90 °C. The reaction mixture was diluted with water (80 mL), and the aqueous phase was extracted with dichloromethane (60 mL) three times. The combined organic layers were washed with brine, dried over sodium sulfate, filtered, and concentrated *in vacuo*. The resulting crude material was purified by silica gel chromatography (10 g column; eluting with dichloromethane/ methanol/ 0.1% triethylamine; ratio). Concentration *in vacuo* resulted in tert-butyl 3-(4-amino-7-methyl-5-(4-((6-methylpyridin-2-yl)oxy)phenyl)-7H-pyrrolo[2,3-d]pyrimidin-6-yl)pyrrolidine-1-carboxylate (1.1 g, 58%) as a light brown solid.

7-methyl-5-(4-((6-methylpyridin-2-yl)oxy)phenyl)-6-(pyrrolidin-3-yl)-7H-pyrrolo[2,3-d]pyrimidin-4-amine



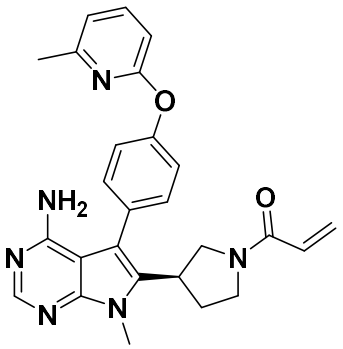
A round bottomed flask was charged with tert-butyl 3-(4-amino-7-methyl-5-(4-((6-methylpyridin-2-yl)oxy)phenyl)-7H-pyrrolo[2,3-d]pyrimidin-6-yl)pyrrolidine-1-carboxylate (1 g, 2 mmol), and a stirbar. TFA/DCM (2/8 mL) was added, and the solution was stirred 30 min at r.t. The reaction mixture was concentrated *in vacuo*. Then dissolved in saturated $NaHCO_3$, extracted with DCM for three times. The combined organic layers were dried with anhydrous Na_2SO_4. Filtered and evaporated to give 7-methyl-5-(4-((6-methylpyridin-2-yl)oxy)phenyl)-6-(pyrrolidin-3-yl)-7H-pyrrolo[2,3-d]pyrimidin-4-amine (700 mg, 87.5%) as an yellow solid.

1-(3-(4-amino-7-methyl-5-(4-((6-methylpyridin-2-yl)oxy)phenyl)-7H-pyrrolo[2,3-d]pyrimidin-6-yl)pyrrolidin-1-yl)prop-2-en-1-one

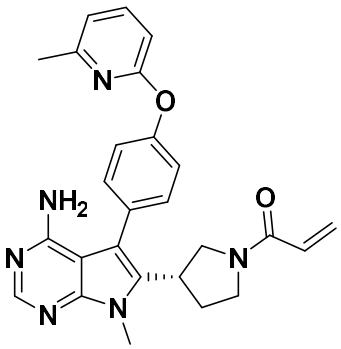


A round bottomed flask was charged with 7-methyl-5-(4-((6-methylpyridin-2-yl)oxy)phenyl)-6-(pyrrolidin-3-yl)-7H-pyrrolo[2,3-d]pyrimidin-4-amine (600 mg, 1.5 mmol), TEA (454.8 mg, 4.49 mmol), dichloromethane (20 mL) and a stirbar. prop-2-enoyl chloride (135.6 mg, 1.5 mmol) was added at −30 °C, and the solution was stirred for 30 min at −30 °C. The reaction mixture was filtered through a pad of Celite, the pad was washed with DCM, and the filtrate was concentrated *in vacuo*. The resulting crude material was purified by HPLC (acetonitrile/water/0.1% formic acid). Lyophilization yielded 1-(3-(4-amino-7-methyl-5-(4-((6-methylpyridin-2-yl)oxy)phenyl)-7H-pyrrolo[2,3-d]pyrimidin-6-yl)pyrrolidin-1-yl)prop-2-en-1-one (300 mg, 44.0%) as an off-white amorphous solid.

(R)-1-(3-(4-amino-7-methyl-5-(4-((6-methylpyridin-2-yl)oxy)phenyl)-7H-pyrrolo[2,3-d]pyrimidin-6-yl)pyrrolidin-1-yl)prop-2-en-1-one (**7**) and (S)-1-(3-(4-amino-7-methyl-5-(4-((6-methylpyridin-2-yl)oxy)phenyl)-7H-pyrrolo[2,3-d]pyrimidin-6-yl)pyrrolidin-1-yl)prop-2-en-1-one (**8**)



7

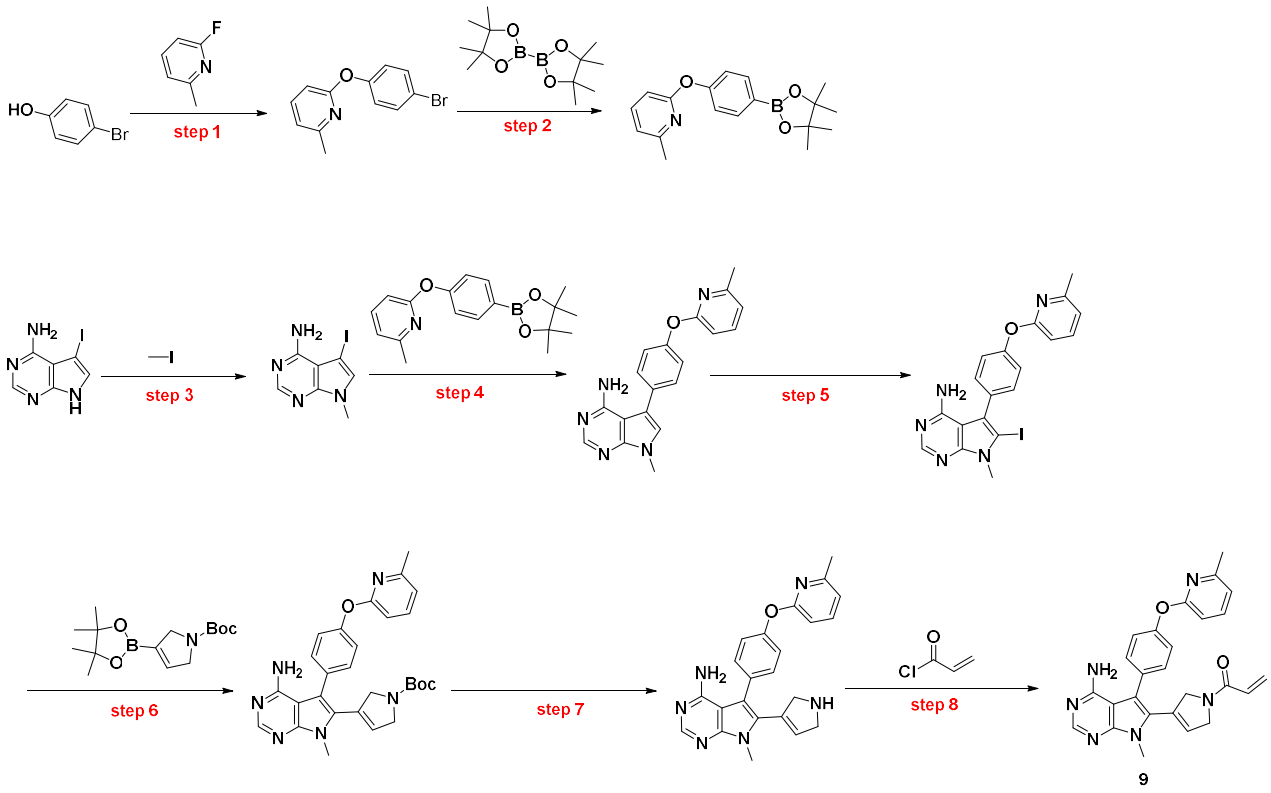


8

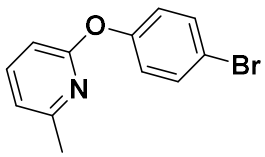
The material 1-(3-(4-amino-7-methyl-5-(4-((6-methylpyridin-2-yl)oxy)phenyl)-7H-pyrrolo[2,3-d]pyrimidin-6-yl)pyrrolidin-1-yl)prop-2-en-1-one (280 mg, 616 µmol) was purified by HPLC

(Column: CHIRALPAK IA, 2*25 cm, 5 μm; Mobile Phase A: MTBE(10 mM NH3-MEOH)--HPLC, Mobile Phase B: EtOH--HPLC; Flow rate: 18 mL/min; Gradient: 15% B to 15% B in 20 min; Wavelength: 220/254 nm; RT1(min): 10.525; RT2(min): 12.125; Sample Solvent: EtOH--HPLC; Injection Volume: 0.5 mL; Number Of Runs: 8). Lyophilization yielded (R)-1-(3-(4-amino-7-methyl-5-(4-((6-methylpyridin-2-yl)oxy)phenyl)-7H-pyrrolo[2,3-d]pyrimidin-6-yl)pyrrolidin-1-yl)prop-2-en-1-one **7** (65 mg, 23.2%) as a white amorphous solid. LC/MS(BAS1): $[M+H]^+$= 455.20; t_R =1.255 min. ^{1}H NMR (400 MHz, DMSO-d_6) δ 8.14 (d, *J* = 1.2 Hz, 1H), 7.85 – 7.69 (m, 1H), 7.50 – 7.32 (m, 2H), 7.20 (dd, *J* = 8.5, 3.8 Hz, 2H), 7.04 (d, *J* = 7.3 Hz, 1H), 6.84 (dd, *J* = 8.1, 4.2 Hz, 1H), 6.49 (ddd, *J* = 33.6, 16.8, 10.3 Hz, 1H), 6.10 (dt, *J* = 16.6, 2.0 Hz, 1H), 5.80 – 5.42 (m, 2H), 3.95 (t, *J* = 9.2 Hz, 1H), 3.80 (d, *J* = 6.5 Hz, 4H), 3.73 – 3.44 (m, 3H), 2.36 (d, *J* = 3.8 Hz, 3H), 2.30 – 1.89 (m, 2H). (S)-1-(3-(4-amino-7-methyl-5-(4-((6-methylpyridin-2-yl)oxy)phenyl)-7H-pyrrolo[2,3-d]pyrimidin-6-yl)pyrrolidin-1-yl)prop-2-en-1-one **8** (62 mg, 22.1%) was isolated as a white amorphous solid. LC/MS(BAS1): $[M+H]^+$= 455.20; t_R =1.257 min. ^{1}H NMR (400 MHz, DMSO-d_6) δ 8.12 (d, *J* = 2.1 Hz, 1H), 7.85 – 7.71 (m, 1H), 7.49 – 7.35 (m, 2H), 7.19 (dd, *J* = 8.4, 4.0 Hz, 2H), 7.04 (d, *J* = 7.3 Hz, 1H), 6.84 (dd, *J* = 8.2, 4.1 Hz, 1H), 6.49 (ddd, *J* = 33.1, 16.8, 10.2 Hz, 1H), 6.10 (dt, *J* = 16.8, 1.9 Hz, 1H), 5.80 (s, 1H), 5.63 (ddd, *J* = 10.9, 8.9, 2.4 Hz, 1H), 3.79 (d, *J* = 6.8 Hz, 4H), 3.73 – 3.44 (m, 3H), 2.36 (d, *J* = 3.8 Hz, 3H), 2.28 – 1.87 (m, 2H).

1-(3-(4-amino-7-methyl-5-(4-((6-methylpyridin-2-yl)oxy)phenyl)-7H-pyrrolo[2,3-d]pyrimidin-6-yl)-2,5-dihydro-1H-pyrrol-1-yl)prop-2-en-1-one (9)

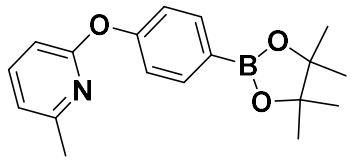


2-(4-bromophenoxy)-6-methylpyridine



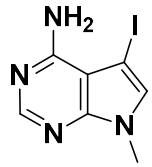
A round bottomed flask was charged with 4-bromo-2-fluorophenol (1.0 g, 5.24 mmol), 2-fluoro-6-methylpyridine (704 mg, 6.28 mmol), Cs_2CO_3 (5.12 g, 15.7 mmol) and a stirbar. DMF (20 mL) was added, and the solution was stirred for 1 h at 130 ºC. The reaction mixture was diluted with H_2O (100 mL), and the aqueous phase was extracted with ethyl acetate (100 mL) three times. The combined organic layers were washed with saturated brines, dried over sodium sulfate, filtered, and concentrated in vacuo. The resulting crude material was purified by HPLC. Concentration in vacuo resulted in 2-(4-bromophenoxy)-6-methylpyridine (1.3 g, 85.0%) as an off-white solid.

2-methyl-6-(4-(4,4,5,5-tetramethyl-1,3,2-dioxaborolan-2-yl)phenoxy)pyridine



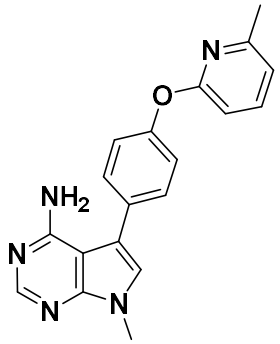
A solution of 2-(4-bromophenoxy)-6-methylpyridine (1.3 g, 4.94 mmol), bis(pinacolato)diboron (1.88 g, 7.41 mmol), KOAc (1.45 g, 14.8 mmol) and $Pd(dppf)Cl_2$ (361 mg, 0.494 mmol) in DMF (20 mL) was stirred for 2 h at 80 ºC under nitrogen atmosphere. The reaction mixture was diluted with H_2O and extracted with ethyl acetate. The combined organic layers were washed with brines, dried over anhydrous Na_2SO_4. After filtration, the filtrate was concentrated under reduced pressure. The residue was purified by HPLC. Concentration in vacuo resulted in 2-methyl-6-(4-(4,4,5,5-tetramethyl-1,3,2-dioxaborolan-2-yl)phenoxy)pyridine (1.1 g, 71.5%) as a yellow solid.

5-iodo-7-methyl-7H-pyrrolo[2,3-d]pyrimidin-4-amine



A round bottomed flask was charged with 5-iodo-7H-pyrrolo[2,3-d]pyrimidin-4-amine (1.0 g, 3.84 mmol), Cs_2CO_3 (1.87 g, 5.76 mmol), DMF (20 mL) and a stirbar. Iodomethane (600 mg, 4.22 mmol) was added, and the solution was stirred for 1 h at room temperature. The reaction mixture was diluted with H_2O (100 mL), and the aqueous phase was extracted with ethyl acetate (100 mL) three times. The combined organic layers were washed with saturated brines, dried over sodium sulfate, filtered, and concentrated in vacuo. The resulting crude material was purified by HPLC. Concentration in vacuo resulted in 5-iodo-7-methyl-7H-pyrrolo[2,3-d]pyrimidin-4-amine (700 mg, 66.7 %) as a yellow solid.

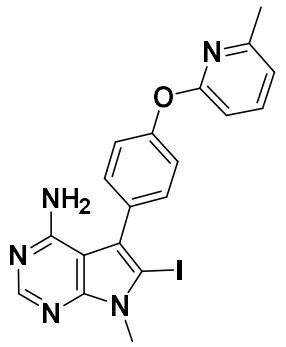
7-methyl-5-(4-((6-methylpyridin-2-yl)oxy)phenyl)-7H-pyrrolo[2,3-d]pyrimidin-4-amine



A resealable reaction vial was charged with 5-iodo-7-methyl-7H-pyrrolo[2,3-d]pyrimidin-4-amine (700 mg, 2.55 mmol), 2-methyl-6-(4-(4,4,5,5-tetramethyl-1,3,2-dioxaborolan-2-

yl)phenoxy)pyridine (953.4 mg, 3.06 mmol), Pd(dppf)Cl_2 (170 mg, 0.225 mmol), K_3PO_4 (1.62 g, 7.65 mmol), DMF (10 mL), H_2O (1.25 mL) and a stir bar before being evacuated and purged with nitrogen three times. The mixture was stirred for 1 h at 90 °C. The reaction mixture was diluted with H_2O (100 mL), and the aqueous phase was extracted with EA (100 mL) three times. The combined organic layers were washed with brines, dried over sodium sulfate, filtered, and concentrated in vacuo. The resulting crude material was purified by HPLC. Concentration in vacuo resulted in 7-methyl-5-(4-((6-methylpyridin-2-yl)oxy)phenyl)-7H-pyrrolo[2,3-d]pyrimidin-4-amine (600.0 mg, 70.9%) as a yellow solid.

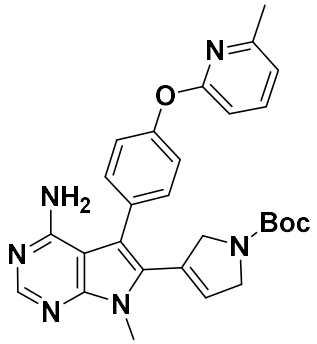
6-iodo-7-methyl-5-(4-((6-methylpyridin-2-yl)oxy)phenyl)-7H-pyrrolo[2,3-d]pyrimidin-4-amine



A round bottomed flask was charged with 7-methyl-5-(4-((6-methylpyridin-2-yl)oxy)phenyl)-7H-pyrrolo[2,3-d]pyrimidin-4-amine (600.0 mg, 1.81 mmol), DCM (20 mL) and TFA (0.5 mL) and a stirbar. 1-iodopyrrolidine-2,5-dione (488.2 mg, 2.17 mmol) was added, and the solution was stirred for 1 h at room temperature. The reaction mixture was diluted with Na_2SO_3 solution (50 mL), and the aqueous phase was extracted with DCM (50 mL) three times. The combined organic layers were washed with saturated brines, dried over sodium sulfate, filtered, and concentrated in vacuo. The resulting crude material was purified by HPLC. Concentration in

vacuo resulted in 6-iodo-7-methyl-5-(4-((6-methylpyridin-2-yl)oxy)phenyl)-7H-pyrrolo[2,3-d]pyrimidin-4-amine (580.5 mg, 70.0 %) as a yellow solid.

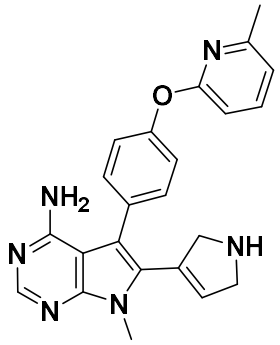
Tert-butyl 3-(4-amino-7-methyl-5-(4-((6-methylpyridin-2-yl)oxy)phenyl)-7H-pyrrolo[2,3-d]pyrimidin-6-yl)-2,5-dihydro-1H-pyrrole-1-carboxylate



A resealable reaction vial was charged with 6-iodo-7-methyl-5-(4-((6-methylpyridin-2-yl)oxy)phenyl)-7H-pyrrolo[2,3-d]pyrimidin-4-amine (85 mg, 0.186 mmol), tert-butyl 3-(4,4,5,5-tetramethyl-1,3,2-dioxaborolan-2-yl)-2,5-dihydro-1H-pyrrole-1-carboxylate (65.8 mg, 0.223 mmol), Pd(dppf)Cl_2 (13.6 mg, 0.018 mmol), K_3PO_4 (118.4 mg, 0.558 mmol), DMF (2 mL), H_2O (0.25 mL) and a stir bar before being evacuated and purged with nitrogen three times. The mixture was stirred for 1 h at 90 °C. The reaction mixture was diluted with H_2O (10 mL), and the aqueous phase was extracted with DCM (10 mL) three times. The combined organic layers were washed with brines, dried over sodium sulfate, filtered, and concentrated in vacuo. The resulting crude material was purified by silica gel chromatography (eluting with MeOH/DCM=1/40). Concentration in vacuo resulted in tert-butyl 3-(4-amino-7-methyl-5-(4-((6-methylpyridin-2-

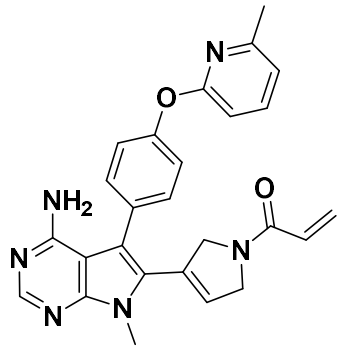
yl)oxy)phenyl)-7H-pyrrolo[2,3-d]pyrimidin-6-yl)-2,5-dihydro-1H-pyrrole-1-carboxylate (50 mg, 53.9%) as a yellow solid.

6-(2,5-dihydro-1H-pyrrol-3-yl)-7-methyl-5-(4-((6-methylpyridin-2-yl)oxy)phenyl)-7H-pyrrolo[2,3-d]pyrimidin-4-amine



A round bottomed flask was charged with tert-butyl 3-(4-amino-7-methyl-5-(4-((6-methylpyridin-2-yl)oxy)phenyl)-7H-pyrrolo[2,3-d]pyrimidin-6-yl)-2,5-dihydro-1H-pyrrole-1-carboxylate (50 mg, 0.1 mmol), DCM (2 mL) and a stir bar. TFA (0.5 mL) was added. The reaction mixture was stirred for 1 h at room temperature. The solvent was removed in vacuo to result in 6-(2,5-dihydro-1H-pyrrol-3-yl)-7-methyl-5-(4-((6-methylpyridin-2-yl)oxy)phenyl)-7H-pyrrolo[2,3-d]pyrimidin-4-amine (40 mg, 100%) as a dark oil.

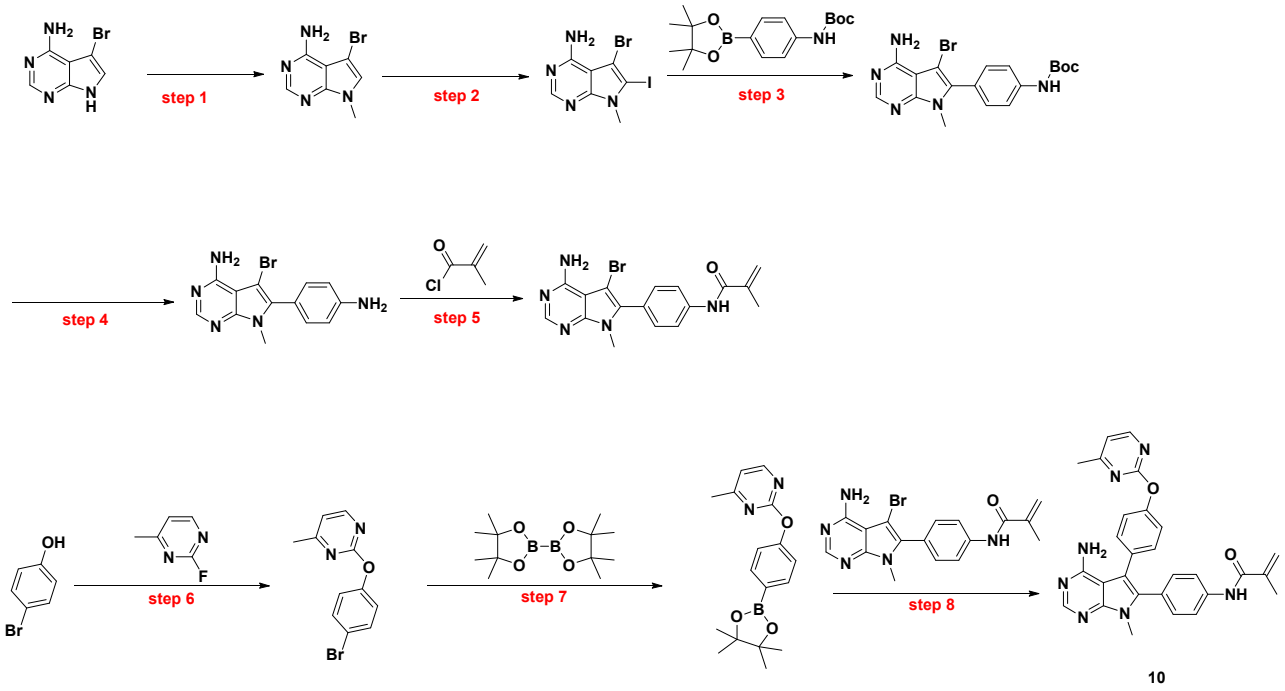
1-(3-(4-amino-7-methyl-5-(4-((6-methylpyridin-2-yl)oxy)phenyl)-7H-pyrrolo[2,3-d]pyrimidin-6-yl)-2,5-dihydro-1H-pyrrol-1-yl)prop-2-en-1-one (**9**)



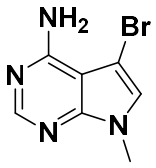
A round bottomed flask was charged with 6-(2,5-dihydro-1H-pyrrol-3-yl)-7-methyl-5-(4-((6-methylpyridin-2-yl)oxy)phenyl)-7H-pyrrolo[2,3-d]pyrimidin-4-amine (35 mg, 0.088 mmol), Et_3N (26.7 mg, 0.26 mmol), DCM (2 mL) and a stir bar. The mixture was cooled to −15 °C, prop-2-enoyl chloride (7.95 mg, 0.088 mmol) was added dropwise and the solution was stirred for 1 h at −15 °C. The reaction mixture was quenched with MeOH, and concentrated in vacuo. The resulting crude material was purified by pre-HPLC. (Column: XBridge Prep OBD C18 Column, 30*150 mm, 5μm; Flow rate: 60 mL/min; Gradient: 20% B to 50% B in 8 min, 50% B; Wavelength: 220 nm; RT1(min): 7.03;). Lyophilization yielded 1-(3-(4-amino-7-methyl-5-(4-((6-methylpyridin-2-yl)oxy)phenyl)-7H-pyrrolo[2,3-d]pyrimidin-6-yl)-2,5-dihydro-1H-pyrrol-1-yl)prop-2-en-1-one **9** (12.3 mg, 30.9 %) as a white amorphous solid. LC/MS(BAS1): $[M+H]^+$= 453.30; t_R =1.342 min. ^{1}H NMR (400 MHz, Chloroform-*d*) δ 8.36 (d, *J* = 3.2 Hz, 1H), 7.70 – 7.59 (m, 1H), 7.40 (dd, *J* = 11.3, 8.3 Hz, 2H), 7.23 (dd, *J* = 11.3, 8.2 Hz, 2H), 6.96 (d, *J* = 7.4 Hz, 1H), 6.79 – 6.70 (m, 1H), 6.47 – 6.37 (m, 1H), 6.24 – 6.11 (m, 1H), 5.79 – 5.64 (m, 1H),

5.02 (s, 2H), 4.52 (s, 2H), 4.32 (s, 1H), 4.22 (s, 1H), 3.88 (d, *J* = 10.2 Hz, 3H), 2.49 (d, *J* = 9.9 Hz, 3H).

N-(4-(4-amino-7-methyl-5-(4-((4-methylpyrimidin-2-yl)oxy)phenyl)-7H-pyrrolo[2,3-d]pyrimidin-6-yl)phenyl)methacrylamide 10

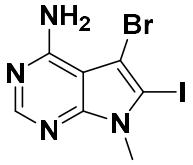


5-bromo-7-methyl-7H-pyrrolo[2,3-d]pyrimidin-4-amine



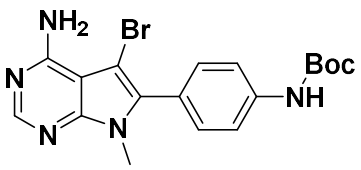
A round bottomed flask was charged with 5-bromo-7H-pyrrolo[2,3-d]pyrimidin-4-amine (10.0 g, 47.16 mmol), Cs_2CO_3 (22.99 g, 70.75 mmol), DMF (120 mL) and a stirbar. Iodomethane (8.03 g, 56.59 mmol) was added, and the solution was stirred for 1 h at room temperature. The reaction mixture was diluted with H_2O (300 mL), and the aqueous phase was extracted with ethyl acetate (300 mL) three times. The combined organic layers were washed with saturated brines, dried over sodium sulfate, filtered, and concentrated in vacuo. The resulting crude material was purified by HPLC. Concentration in vacuo resulted in 5-bromo-7-methyl-7H-pyrrolo[2,3-d]pyrimidin-4-amine (5 g, 47.1 %) as an off-white solid.

5-bromo-6-iodo-7-methyl-7H-pyrrolo[2,3-d]pyrimidin-4-amine



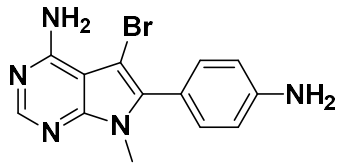
A round bottomed flask was charged with 5-bromo-7-methyl-7H-pyrrolo[2,3-d]pyrimidin-4-amine (5 g, 22.12 mmol), DCM (50 mL) and TFA (2 mL) and a stirbar. 1-iodopyrrolidine-2,5-dione (5.97 g, 26.54 mmol) was added, and the solution was stirred for 2 h at room temperature. The reaction mixture was diluted with Na_2SO_3 solution (200 mL), and the aqueous phase was extracted with DCM (200 mL) three times. The combined organic layers were washed with saturated brines, dried over sodium sulfate, filtered, and concentrated in vacuo. The resulting crude material was purified by HPLC. Concentration in vacuo resulted in 5-bromo-6-iodo-7-methyl-7H-pyrrolo[2,3-d]pyrimidin-4-amine (4 g, 51.3 %) as a yellow solid.

tert-butyl (4-(4-amino-5-bromo-7-methyl-7H-pyrrolo[2,3-d]pyrimidin-6-yl)phenyl)carbamate



A resealable reaction vial was charged with 5-bromo-6-iodo-7-methyl-7H-pyrrolo[2,3-d]pyrimidin-4-amine (4 g, 11.36 mmol), tert-butyl (4-(4,4,5,5-tetramethyl-1,3,2-dioxaborolan-2-yl)phenyl)carbamate (4.35 g, 13.63 mmol), Pd(dppf)Cl_2 (994.16 mg, 1.36 mmol), K_3PO_4 (7.22 g, 34.08 mmol), DMF (50 mL), H_2O (6.25 mL) and a stir bar before being evacuated and purged with nitrogen three times. The mixture was stirred for 2 h at 90 °C. The reaction mixture was diluted with H_2O (300 mL), and the aqueous phase was extracted with EA (300 mL) three times. The combined organic layers were washed with brines, dried over sodium sulfate, filtered, and concentrated in vacuo. The resulting crude material was purified by silica gel chromatography (eluting with MeOH/DCM = 1/40). Concentration in vacuo resulted in tert-butyl (4-(4-amino-5-bromo-7-methyl-7H-pyrrolo[2,3-d]pyrimidin-6-yl)phenyl)carbamate (3 g, 63.4%) as a yellow solid.

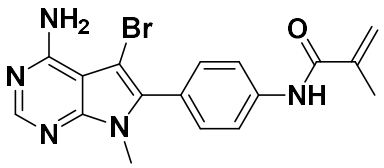
6-(4-aminophenyl)-5-bromo-7-methyl-7H-pyrrolo[2,3-d]pyrimidin-4-amine



A round bottomed flask was charged with tert-butyl (4-(4-amino-5-bromo-7-methyl-7H-pyrrolo[2,3-d]pyrimidin-6-yl)phenyl)carbamate (3 g, 7.19 mmol), DCM (50 mL) and TFA (12.5

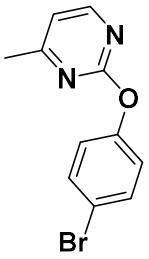
mL) and a stirbar. The solution was stirred for 1 h at room temperature. The reaction mixture was diluted with H2O (100 mL), and the aqueous phase was extracted with DCM (50 mL) three times. The pH of aqueous phase was adjusted to 7~8, then the aqueous phase was extracted with DCM (100 mL) three times. The combined organic layers were washed with saturated brines, dried over sodium sulfate, filtered, and concentrated in vacuo resulted in 6-(4-aminophenyl)-5-bromo-7-methyl-7H-pyrrolo[2,3-d]pyrimidin-4-amine (2.1 g, 92.1%) as a yellow solid.

N-(4-(4-amino-5-bromo-7-methyl-7H-pyrrolo[2,3-d]pyrimidin-6-yl)phenyl)methacrylamide



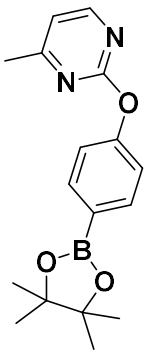
A resealable reaction vial was charged with 6-(4-aminophenyl)-5-bromo-7-methyl-7H-pyrrolo[2,3-d]pyrimidin-4-amine (2.1 g, 6.62 mmol), pyridine (785 mg, 9.93 mmol), DCM (100 mL) and a stir bar before being evacuated and purged with nitrogen three times. Methacryloyl chloride (757.3 mg, 7.28 mmol) was added slowly at 0 ºC. Then the mixture was stirred for 2 h at room temperature. The reaction mixture was diluted with H_2O (100 mL), and the aqueous phase was extracted with DCM (100 mL) three times. The combined organic layers were washed with brines, dried over sodium sulfate, filtered, and concentrated in vacuo. The resulting crude material was purified by silica gel chromatography (eluting with MeOH/DCM = 1/40). Concentration in vacuo resulted in N-(4-(4-amino-5-bromo-7-methyl-7H-pyrrolo[2,3-d]pyrimidin-6-yl)phenyl)methacrylamide (1.8 g, 70.5%) as an off-white solid.

2-(4-bromophenoxy)-4-methylpyrimidine



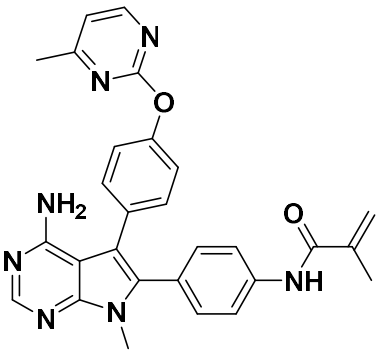
A round bottomed flask was charged with 4-bromophenol (1.0 g, 5.78 mmol), 2-fluoro-4-methylpyrimidine (777.6 mg, 6.94 mmol), Cs_2CO_3 (5.65 g, 17.34 mmol) and a stirbar. DMF (20 mL) was added, and the solution was stirred for 1 h at 100 ºC. The reaction mixture was diluted with H_2O (100 mL), and the aqueous phase was extracted with ethyl acetate (100 mL) three times. The combined organic layers were washed with saturated brines, dried over sodium sulfate, filtered, and concentrated in vacuo. The resulting crude material was purified by HPLC. Concentration in vacuo resulted in 2-(4-bromophenoxy)-4-methylpyrimidine (1.4 g, 91.3 %) as a yellow amorphous solid.

4-methyl-2-(4-(4,4,5,5-tetramethyl-1,3,2-dioxaborolan-2-yl)phenoxy)pyrimidine



A solution of 2-(4-bromophenoxy)-4-methylpyrimidine (500.00 mg, 1.89 mmol), bis(pinacolato)diboron (718.4 mg, 2.83 mmol), KOAc (555 mg, 5.66 mmol) and Pd(dppf)Cl_2 (138 mg, 0.189 mmol) in DMF (10 mL) was stirred for 2 h at 80 ºC under nitrogen atmosphere. The resulting mixture was extracted with EA. The combined organic layers were washed with brines, dried over anhydrous Na_2SO_4. After filtration, the filtrate was concentrated under reduced pressure. The residue was purified by HPLC. Concentration in vacuo resulted in 4-methyl-2-(4-(4,4,5,5-tetramethyl-1,3,2-dioxaborolan-2-yl)phenoxy)pyrimidine (500 mg, 84.9%) as a yellow solid.

N-(4-(4-amino-7-methyl-5-(4-((4-methylpyrimidin-2-yl)oxy)phenyl)-7H-pyrrolo[2,3-d]pyrimidin-6-yl)phenyl)methacrylamide (**10**)

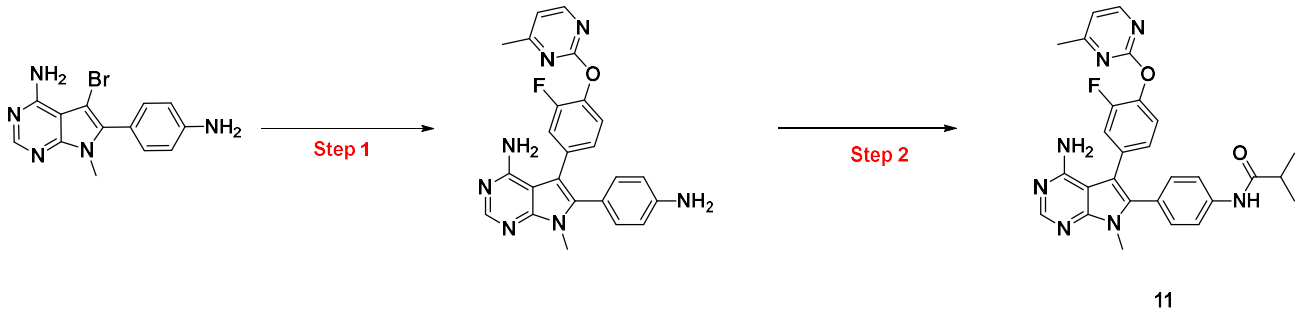


A resealable reaction vial was charged with N-(4-(4-amino-5-bromo-7-methyl-7H-pyrrolo[2,3-d]pyrimidin-6-yl)phenyl)methacrylamide (100 mg, 0.259 mmol), 4-methyl-2-(4-(4,4,5,5-tetramethyl-1,3,2-dioxaborolan-2-yl)phenoxy)pyrimidine (97 mg, 0.31 mmol), Pd(DtBPF)Cl_2 (16.9 mg, 0.026 mmol), CsF (118 mg, 0.776 mmol), DMF (2 mL), H_2O (0.25 mL) and a stir bar before being evacuated and purged with nitrogen three times. The mixture was stirred for 1 h at 90 °C. The reaction mixture was diluted with H_2O (10 mL), and the aqueous phase was extracted with DCM (10 mL) three times. The combined organic layers were washed with brines, dried

over sodium sulfate, filtered, and concentrated in vacuo. The resulting crude material was purified by HPLC (Column: XBridge Prep C18 OBD Column, 19*150 mm, 5 μm; Mobile Phase A: Water(10MMOL/L NH_4HCO_3+0.1%$NH_3.H_2O$), Mobile Phase B: DCM: EtOH=9: 1--HPLC; Flow rate: 25 mL/min; Gradient: 45% B to 65% B in 7 min, 65% B; Wavelength: 254/220 nm; RT1(min): 6.5). Concentration in vacuo resulted in N-(4-(4-amino-7-methyl-5-(4-((4-methylpyrimidin-2-yl)oxy)phenyl)-7H-pyrrolo[2,3-d]pyrimidin-6-yl)phenyl)methacrylamide **10** (18 mg, 14.1%) as an off-white solid. LC/MS(BAS1):$[M+H]^+$= 492.20; t_R =1.365 min. ^{1}H NMR (400 MHz, DMSO-d_6) δ 9.91 (s, 1H), 8.47 (d, J = 5.0 Hz, 1H), 8.20 (s, 1H), 7.77 – 7.69 (m, 2H), 7.35 – 7.25 (m, 4H), 7.22 – 7.12 (m, 3H), 5.91 (s, 1H), 5.80 (t, J = 1.0 Hz, 1H), 5.56 – 5.51 (m, 1H), 3.61 (s, 3H), 2.42 (s, 3H), 1.95 (t, J = 1.2 Hz, 3H).

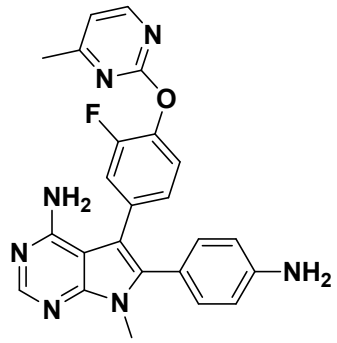
N-(4-(4-amino-5-(3-fluoro-4-((4-methylpyrimidin-2-yl)oxy)phenyl)-7-methyl-7H-pyrrolo[2,3-d]pyrimidin-6-yl)phenyl)isobutyramide

(11)



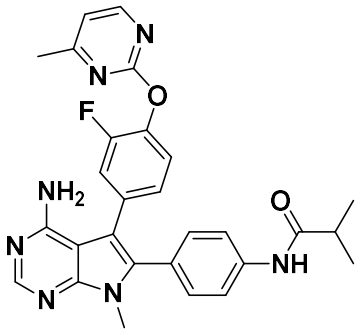
11

6-(4-aminophenyl)-5-(3-fluoro-4-((4-methylpyrimidin-2-yl)oxy)phenyl)-7-methyl-7H-pyrrolo[2,3-d]pyrimidin-4-amine



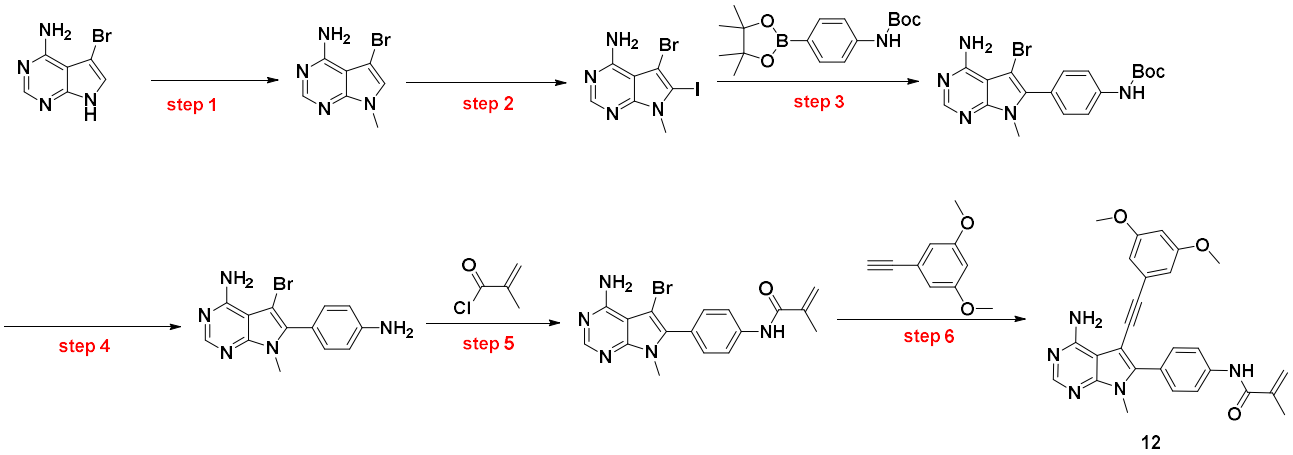
A round-bottomed flask was charged with 6-(4-aminophenyl)-5-bromo-7-methyl-7H-pyrrolo[2,3-d]pyrimidin-4-amine (4 g, 12.57 mmol), 2-(2-fluoro-4-(4,4,5,5-tetramethyl-1,3,2-dioxaborolan-2-yl)phenoxy)-4-methylpyrimidine (4.98 g, 15.08 mmol), $Pd(dppf)Cl_2$ (920 mg, 1.26 mmol), K3PO4 (8 g, 37.71 mmol), DMF (80 mL), H_2O (10 mL) and a stir bar before being evacuated and purged with nitrogen three times. The mixture was stirred for 2 h at 90 °C. The reaction mixture was diluted with H_2O (100 mL), and the aqueous phase was extracted with EtOAc (100 mL) three times. The combined organic layers were washed with brines, dried over sodium sulfate, filtered, and concentrated in vacuo. The resulting crude material was purified by silica gel column. Concentration in vacuo resulted in 6-(4-aminophenyl)-5-(3-fluoro-4-((4-methylpyrimidin-2-yl)oxy)phenyl)-7-methyl-7H-pyrrolo[2,3-d]pyrimidin-4-amine (1.2 g, 21.6%) as an off-white solid.

N-(4-(4-amino-5-(3-fluoro-4-((4-methylpyrimidin-2-yl)oxy)phenyl)-7-methyl-7H-pyrrolo[2,3-d]pyrimidin-6-yl)phenyl)isobutyramide (**11**)

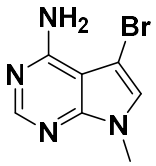


A mixture of 6-(4-aminophenyl)-5-{3-fluoro-4-[(4-methylpyrimidin-2-yl)oxy]phenyl}-7-methyl-7H-pyrrolo[2,3-d]pyrimidin-4-amine (250 mg, 566 μmol) in TFE (3 ml) was added 2-methylpropanoyl 2-methylpropanoate (178 mg, 2 eq) in TFE (1.5 mL) dropwise at 20°C. The solution was stirred at r.t. for 6h and purified by Prep-HPLC with the following conditions: Column: XBridge Prep OBD C18 Column, 30×150mm 5um; Mobile Phase A: Water (10MMOL/L NH_4HCO_3+0.1%$NH_3.H_2O$), Mobile Phase B: ACN; Flow rate: 60 mL/min; Gradient:25 B to 55 B in 7 min, 220 nm; RT1:5.75. This resulted in N-[4-(4-amino-5-{3-fluoro-4-[(4-methylpyrimidin-2-yl)oxy]phenyl}-7-methyl-7H-pyrrolo[2,3-d]pyrimidin-6-yl)phenyl]-2-methylpropanamide 11 (200 mg, 390 μmol, 69.2 %) as a white solid. m/z (ES^+) $[M+H]^+$ = 512.40; HPLC tR = 0.855 min. ^1H NMR (400 MHz, DMSO-d_6) δ 9.99 (s, 1H), 8.47 (d, J = 5.0 Hz, 1H), 8.21 (s, 1H), 7.68 (d, J = 8.4 Hz, 2H), 7.38 - 7.29 (m, 2H), 7.29 (s, 1H), 7.22 - 7.14 (m, 2H), 7.10 (dd, J = 8.2, 2.1 Hz, 1H), 6.00 (s, 2H), 3.58 (s, 3H), 2.60 (p, J = 6.7 Hz, 1H), 2.41 (s, 3H), 1.10 (d, J = 6.8 Hz, 6H).

N-(4-(4-amino-5-((3,5-dimethoxyphenyl)ethynyl)-7-methyl-7H-pyrrolo[2,3-d]pyrimidin-6-yl)phenyl)methacrylamide (12)



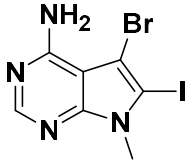
5-bromo-7-methyl-7H-pyrrolo[2,3-d]pyrimidin-4-amine



A round bottomed flask was charged with 5-bromo-7H-pyrrolo[2,3-d]pyrimidin-4-amine (10.0 g, 47.16 mmol), Cs_2CO_3 (22.99 g, 70.75 mmol), DMF (120 mL) and a stirbar. Iodomethane (8.03 g, 56.59 mmol) was added, and the solution was stirred for 1 h at room temperature. The reaction mixture was diluted with H_2O (300 mL), and the aqueous phase was extracted with ethyl acetate (300 mL) three times. The combined organic layers were washed with saturated brines, dried over sodium sulfate, filtered, and concentrated in vacuo. The resulting crude material was

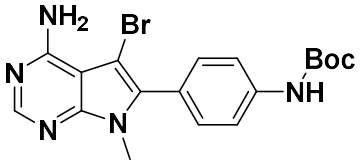
purified by HPLC. Concentration in vacuo resulted in 5-bromo-7-methyl-7H-pyrrolo[2,3-d]pyrimidin-4-amine (5 g, 47.1%) as an off-white solid.

5-bromo-6-iodo-7-methyl-7H-pyrrolo[2,3-d]pyrimidin-4-amine



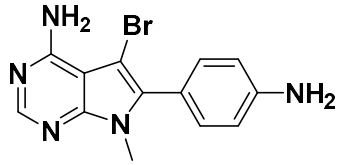
A round bottomed flask was charged with 5-bromo-7-methyl-7H-pyrrolo[2,3-d]pyrimidin-4-amine (5 g, 22.12 mmol), DCM (50 mL) and TFA (2 mL) and a stirbar. 1-iodopyrrolidine-2,5-dione (5.97 g, 26.54 mmol) was added, and the solution was stirred for 2 h at room temperature. The reaction mixture was diluted with Na_2SO_3 solution (200 mL), and the aqueous phase was extracted with DCM (200 mL) three times. The combined organic layers were washed with saturated brines, dried over sodium sulfate, filtered, and concentrated in vacuo. The resulting crude material was purified by HPLC. Concentration in vacuo resulted in 5-bromo-6-iodo-7-methyl-7H-pyrrolo[2,3-d]pyrimidin-4-amine (4 g, 51.3%) as a yellow solid.

tert-butyl (4-(4-amino-5-bromo-7-methyl-7H-pyrrolo[2,3-d]pyrimidin-6-yl)phenyl)carbamate



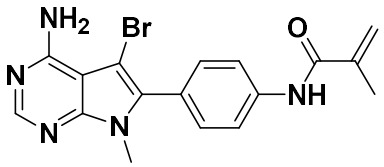
A resealable reaction vial was charged with 5-bromo-6-iodo-7-methyl-7H-pyrrolo[2,3-d]pyrimidin-4-amine (4 g, 11.36 mmol), tert-butyl (4-(4,4,5,5-tetramethyl-1,3,2-dioxaborolan-2-yl)phenyl)carbamate (4.35 g, 13.63 mmol), Pd(dppf)Cl_2 (994.16 mg, 1.36 mmol), K_3PO_4 (7.22 g, 34.08 mmol), DMF (50 mL), H_2O (6.25 mL) and a stir bar before being evacuated and purged with nitrogen three times. The mixture was stirred for 2 h at 90 °C. The reaction mixture was diluted with H_2O (300 mL), and the aqueous phase was extracted with EA (300 mL) three times. The combined organic layers were washed with brines, dried over sodium sulfate, filtered, and concentrated in vacuo. The resulting crude material was purified by silica gel chromatography (eluting with MeOH/DCM=1/40). Concentration in vacuo resulted in tert-butyl (4-(4-amino-5-bromo-7-methyl-7H-pyrrolo[2,3-d]pyrimidin-6-yl)phenyl)carbamate (3 g, 63.4%) as a yellow solid.

6-(4-aminophenyl)-5-bromo-7-methyl-7H-pyrrolo[2,3-d]pyrimidin-4-amine



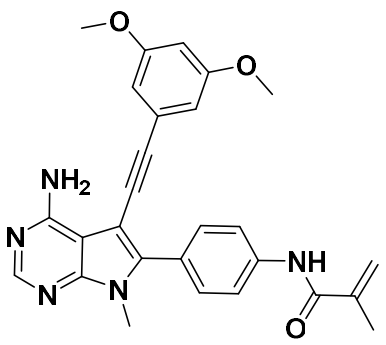
A round bottomed flask was charged with tert-butyl (4-(4-amino-5-bromo-7-methyl-7H-pyrrolo[2,3-d]pyrimidin-6-yl)phenyl)carbamate (3 g, 7.19 mmol), DCM (50 mL) and TFA (12.5 mL) and a stirbar. The solution was stirred for 1 h at room temperature. The reaction mixture was diluted with H_2O (100 mL), and the aqueous phase was extracted with DCM (50 mL) three times. The pH of aqueous phase was adjusted to 7~8, then the aqueous phase was extracted with DCM (100 mL) three times. The combined organic layers were washed with saturated brines, dried over sodium sulfate, filtered, and concentrated in vacuo resulted in 6-(4-aminophenyl)-5-bromo-7-methyl-7H-pyrrolo[2,3-d]pyrimidin-4-amine (2.1 g, 92.1 %) as a yellow solid.

N-(4-(4-amino-5-bromo-7-methyl-7H-pyrrolo[2,3-d]pyrimidin-6-yl)phenyl)methacrylamide



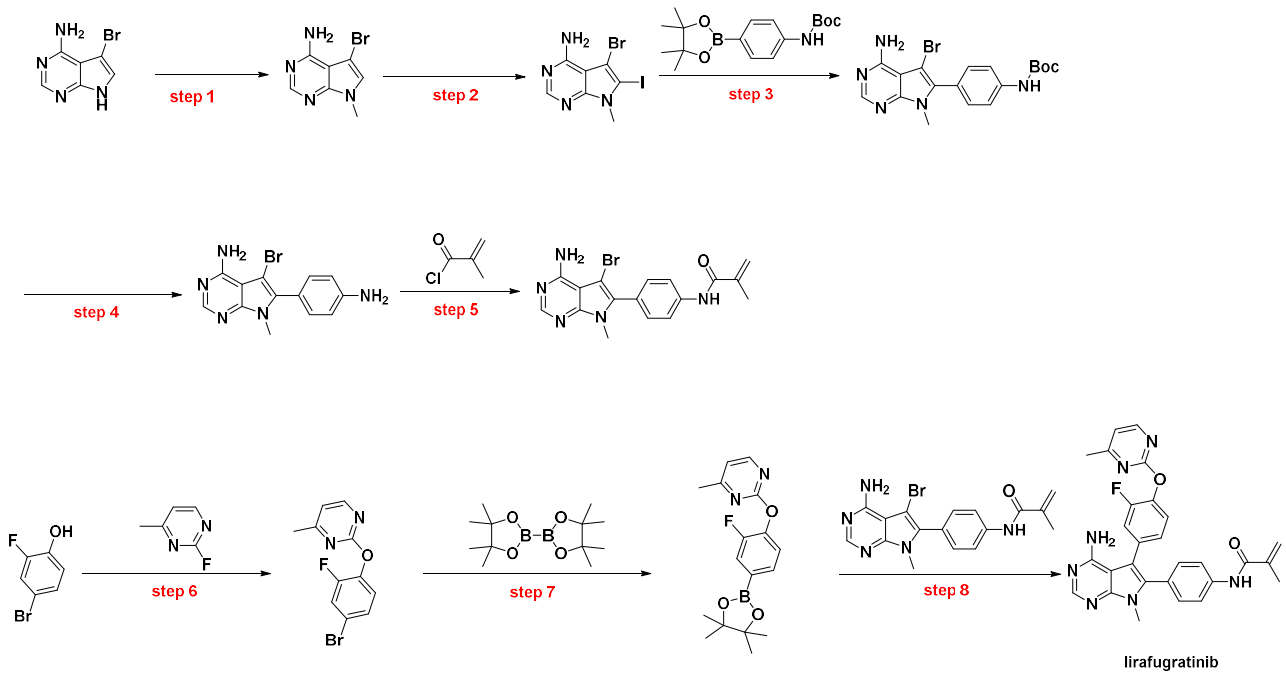
A resealable reaction vial was charged with 6-(4-aminophenyl)-5-bromo-7-methyl-7H-pyrrolo[2,3-d]pyrimidin-4-amine (2.1 g, 6.62 mmol), pyridine (785 mg, 9.93 mmol), DCM (100 mL) and a stir bar before being evacuated and purged with nitrogen three times. Methacryloyl chloride (757.3 mg, 7.28 mmol) was added slowly at 0 ºC. Then the mixture was stirred for 2 h at room temperature. The reaction mixture was diluted with H_2O (100 mL), and the aqueous phase was extracted with DCM (100 mL) three times. The combined organic layers were washed with brines, dried over sodium sulfate, filtered, and concentrated in vacuo. The resulting crude material was purified by silica gel chromatography (eluting with MeOH/DCM=1/40). Concentration in vacuo resulted in N-(4-(4-amino-5-bromo-7-methyl-7H-pyrrolo[2,3-d]pyrimidin-6-yl)phenyl)methacrylamide (1.8 g, 70.5%) as an off-white solid.

N-(4-(4-amino-5-((3,5-dimethoxyphenyl)ethynyl)-7-methyl-7H-pyrrolo[2,3-d]pyrimidin-6-yl)phenyl)methacrylamide (**12**)

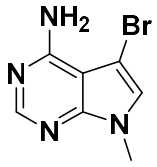


A resealable reaction vial was charged with N-(4-(4-amino-5-bromo-7-methyl-7H-pyrrolo[2,3-d]pyrimidin-6-yl)phenyl)methacrylamide (300 mg, 0.77 mmol), 1-ethynyl-3,5-dimethoxybenzene (189 mg, 1.17 mmol), $Pd(PPh_3)_2Cl_2$ (54 mg, 0.077 mmol), CuI (59.2 mg, 0.31 mmol), DMF (5 mL), TEA (393 mg, 3.88 mmol) and a stir bar before being evacuated and purged with nitrogen three times. The mixture was stirred for 3 h at 100 °C. The reaction mixture was diluted with H_2O (30 mL), and the aqueous phase was extracted with DCM (30 mL) three times. The combined organic layers were washed with brines, dried over sodium sulfate, filtered, and concentrated in vacuo. The resulting crude material was purified by HPLC (Column: Xselect CSH OBD Column 30*150 mm 5 um, n; Mobile Phase A: Water (0.1% FA), Mobile Phase B: ACN; Flow rate: 60 mL/min; Gradient: 20% B to 50% B in 7 min, 50% B; Wavelength: 220 nm; RT1(min): 7.10). Concentration in vacuo resulted in N-(4-(4-amino-5-((3,5-dimethoxyphenyl)ethynyl)-7-methyl-7H-pyrrolo[2,3-d]pyrimidin-6-yl)phenyl)methacrylamide **12** (37.4 mg, 10.3%) as an off-white solid. LC/MS(BAS1):$[M+H]^+$= 468.15; t_R =0.897 min. ^{1}H NMR (400 MHz, DMSO-d_6) δ 10.03 (s, 1H), 8.21 (s, 1H), 7.96 – 7.89 (m, 2H), 7.75 – 7.69 (m, 2H), 6.63 (d, J = 2.3 Hz, 3H), 6.52 (t, J = 2.3 Hz, 1H), 5.85 (s, 1H), 5.57 (d, J = 1.6 Hz, 1H), 3.74 (d, J = 17.3 Hz, 9H), 1.99 (d, J = 1.2 Hz, 3H).

N-(4-(4-amino-5-(3-fluoro-4-((4-methylpyrimidin-2-yl)oxy)phenyl)-7-methyl-7H-pyrrolo[2,3-d]pyrimidin-6-yl)phenyl)methacrylamide (lirafugratinib)



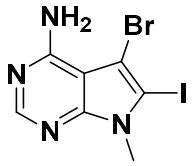
5-bromo-7-methyl-7H-pyrrolo[2,3-d]pyrimidin-4-amine



A round bottomed flask was charged with 5-bromo-7H-pyrrolo[2,3-d]pyrimidin-4-amine (10.0 g, 47.16 mmol), Cs_2CO_3 (22.99 g, 70.75 mmol), DMF (120 mL) and a stirbar. Iodomethane (8.03 g, 56.59 mmol) was added, and the solution was stirred for 1 h at room temperature. The reaction

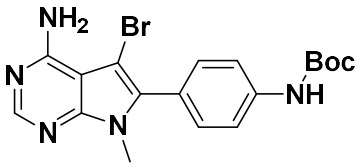
mixture was diluted with H_2O (300 mL), and the aqueous phase was extracted with ethyl acetate (300 mL) three times. The combined organic layers were washed with saturated brines, dried over sodium sulfate, filtered, and concentrated in vacuo. The resulting crude material was purified by HPLC. Concentration in vacuo resulted in 5-bromo-7-methyl-7H-pyrrolo[2,3-d]pyrimidin-4-amine (5 g, 47.1 %) as an off-white solid.

5-bromo-6-iodo-7-methyl-7H-pyrrolo[2,3-d]pyrimidin-4-amine



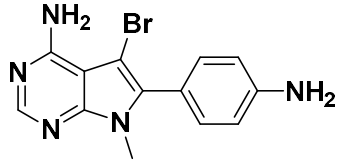
A round bottomed flask was charged with 5-bromo-7-methyl-7H-pyrrolo[2,3-d]pyrimidin-4-amine (5 g, 22.12 mmol), DCM (50 mL) and TFA (2 mL) and a stirbar. 1-iodopyrrolidine-2,5-dione (5.97 g, 26.54 mmol) was added, and the solution was stirred for 2 h at room temperature. The reaction mixture was diluted with Na_2SO_3 solution (200 mL), and the aqueous phase was extracted with DCM (200 mL) three times. The combined organic layers were washed with saturated brines, dried over sodium sulfate, filtered, and concentrated in vacuo. The resulting crude material was purified by HPLC. Concentration in vacuo resulted in 5-bromo-6-iodo-7-methyl-7H-pyrrolo[2,3-d]pyrimidin-4-amine (4 g, 51.3 %) as a yellow solid.

tert-butyl (4-(4-amino-5-bromo-7-methyl-7H-pyrrolo[2,3-d]pyrimidin-6-yl)phenyl)carbamate



A resealable reaction vial was charged with 5-bromo-6-iodo-7-methyl-7H-pyrrolo[2,3-d]pyrimidin-4-amine (4 g, 11.36 mmol), tert-butyl (4-(4,4,5,5-tetramethyl-1,3,2-dioxaborolan-2-yl)phenyl)carbamate (4.35 g, 13.63 mmol), Pd(dppf)Cl_2 (994.16 mg, 1.36 mmol), K_3PO_4 (7.22 g, 34.08 mmol), DMF (50 mL), H_2O (6.25 mL) and a stir bar before being evacuated and purged with nitrogen three times. The mixture was stirred for 2 h at 90 °C. The reaction mixture was diluted with H_2O (300 mL), and the aqueous phase was extracted with ethyl acetate (300 mL) three times. The combined organic layers were washed with brines, dried over sodium sulfate, filtered, and concentrated in vacuo. The resulting crude material was purified by silica gel chromatography (eluting with MeOH/DCM = 1/40). Concentration in vacuo resulted in tert-butyl (4-(4-amino-5-bromo-7-methyl-7H-pyrrolo[2,3-d]pyrimidin-6-yl)phenyl)carbamate (3 g, 63.4%) as a yellow solid.

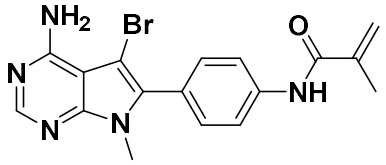
6-(4-aminophenyl)-5-bromo-7-methyl-7H-pyrrolo[2,3-d]pyrimidin-4-amine



A round bottomed flask was charged with tert-butyl (4-(4-amino-5-bromo-7-methyl-7H-pyrrolo[2,3-d]pyrimidin-6-yl)phenyl)carbamate (3 g, 7.19 mmol), DCM (50 mL) and TFA (12.5

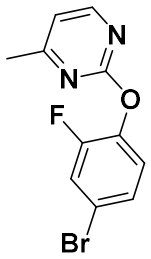
mL) and a stirbar. The solution was stirred for 1 h at room temperature. The reaction mixture was diluted with H_2O (100 mL), and the aqueous phase was extracted with DCM (50 mL) three times. The pH of aqueous phase was adjusted to 7~8, then the aqueous phase was extracted with DCM (100 mL) three times. The combined organic layers were washed with saturated brines, dried over sodium sulfate, filtered, and concentrated in vacuo resulted in 6-(4-aminophenyl)-5-bromo-7-methyl-7H-pyrrolo[2,3-d]pyrimidin-4-amine (2.1 g, 92.1%) as a yellow solid.

N-(4-(4-amino-5-bromo-7-methyl-7H-pyrrolo[2,3-d]pyrimidin-6-yl)phenyl)methacrylamide



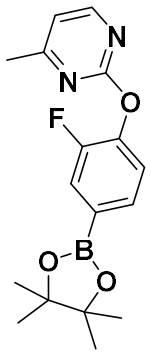
A resealable reaction vial was charged with 6-(4-aminophenyl)-5-bromo-7-methyl-7H-pyrrolo[2,3-d]pyrimidin-4-amine (2.1 g, 6.62 mmol), pyridine (785 mg, 9.93 mmol), DCM (100 mL) and a stir bar before being evacuated and purged with nitrogen three times. Methacryloyl chloride (757.3 mg, 7.28 mmol) was added slowly at 0 ºC. Then the mixture was stirred for 2 h at room temperature. The reaction mixture was diluted with H_2O (100 mL), and the aqueous phase was extracted with DCM (100 mL) three times. The combined organic layers were washed with brines, dried over sodium sulfate, filtered, and concentrated in vacuo. The resulting crude material was purified by silica gel chromatography (eluting with MeOH/DCM=1/40). Concentration in vacuo resulted in N-(4-(4-amino-5-bromo-7-methyl-7H-pyrrolo[2,3-d]pyrimidin-6-yl)phenyl)methacrylamide (1.8 g, 70.5%) as an off-white solid.

2-(4-bromo-2-fluorophenoxy)-4-methylpyrimidine



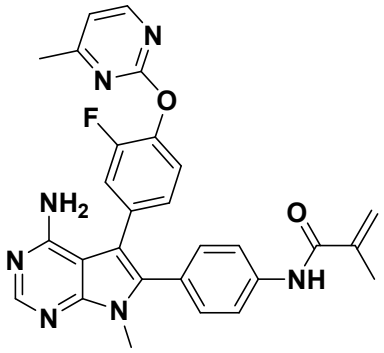
A round bottomed flask was charged with 4-bromo-2-fluorophenol (1.0 g, 5.24 mmol), 2-fluoro-4-methylpyrimidine (704 mg, 6.28 mmol), Cs_2CO_3 (5.12 g, 15.7 mmol) and a stirbar. DMF (20 mL) was added, and the solution was stirred for 1 h at 100 ºC. The reaction mixture was diluted with H_2O (100 mL), and the aqueous phase was extracted with ethyl acetate (100 mL) three times. The combined organic layers were washed with saturated brines, dried over sodium sulfate, filtered, and concentrated in vacuo. The resulting crude material was purified by HPLC. Concentration in vacuo resulted in 2-(4-bromo-2-fluorophenoxy)-4-methylpyrimidine (1.48 g, 99.8 %) as an off-white amorphous solid.

2-(2-fluoro-4-(4,4,5,5-tetramethyl-1,3,2-dioxaborolan-2-yl)phenoxy)-4-methylpyrimidine



A solution/mixture of 2-(4-bromo-2-fluorophenoxy)-4-methylpyrimidine (500.00 mg, 1.77 mmol), bis(pinacolato)diboron (672.75 mg, 2.65 mmol), KOAc (520 mg, 5.3 mmol) and Pd(dppf)Cl_2 (129.4 mg, 0.177 mmol) in DMF (10 mL) was stirred for 2 h at 80 ºC under nitrogen atmosphere. The resulting mixture was diluted with water and extracted with EA. The combined organic layers were washed with brines, dried over anhydrous Na_2SO_4. After filtration, the filtrate was concentrated under reduced pressure. The residue was purified by HPLC. Concentration in vacuo resulted in 2-(2-fluoro-4-(4,4,5,5-tetramethyl-1,3,2-dioxaborolan-2-yl)phenoxy)-4-methylpyrimidine (430 mg, 73.7%) as a yellow solid.

N-(4-(4-amino-5-(3-fluoro-4-((4-methylpyrimidin-2-yl)oxy)phenyl)-7-methyl-7H-pyrrolo[2,3-d]pyrimidin-6-yl)phenyl)methacrylamide (lirafugratinib)



A resealable reaction vial was charged with N-(4-(4-amino-5-bromo-7-methyl-7H-pyrrolo[2,3-d]pyrimidin-6-yl)phenyl)methacrylamide (100 mg, 0.259 mmol), 2-(2-fluoro-4-(4,4,5,5-tetramethyl-1,3,2-dioxaborolan-2-yl)phenoxy)-4-methylpyrimidine (102.6 mg, 0.310 mmol), Pd(DtBPF)Cl_2 (16.9 mg, 0.026 mmol), CsF (118 mg, 0.776 mmol), DMF (2 mL), H_2O (0.25 mL) and a stir bar before being evacuated and purged with nitrogen three times. The mixture was stirred for 1 h at 90 °C. The reaction mixture was diluted with H_2O (10 mL), and the aqueous

phase was extracted with DCM (10 mL) three times. The combined organic layers were washed with brines, dried over sodium sulfate, filtered, and concentrated in vacuo. The resulting crude material was purified by Pre-HPLC (Column: XBridge Prep C18 OBD Column, 19*150 mm, 5μm; Mobile Phase A: Water(10 mmol/L NH_4HCO_3), Mobile Phase B: ACN; Flow rate: 25 mL/min; Gradient: 25% B to 50% B in 7 min, 50% B; Wave Length: 254/220 nm; RT1(min): 6.5). Concentration in vacuo resulted in N-(4-(4-amino-5-(3-fluoro-4-((4-methylpyrimidin-2-yl)oxy)phenyl)-7-methyl-7H-pyrrolo[2,3-d]pyrimidin-6-yl)phenyl)methacrylamide (27.2 mg, 20.6%) as an off-white solid. LC/MS(BAS1): $[M+H]^+$= 510.20; t_R =1.405 min. ^{1}H NMR (400 MHz, DMSO-d_6) δ 9.92 (s, 1H), 8.47 (d, *J* = 5.0 Hz, 1H), 8.21 (s, 1H), 7.79 – 7.72 (m, 2H), 7.38 – 7.28 (m, 3H), 7.22 – 7.14 (m, 2H), 7.10 (dd, *J* = 8.1, 2.1 Hz, 1H), 5.98 (s, 2H), 5.80 (s, 1H), 5.54 (d, *J* = 1.7 Hz, 1H), 3.59 (s, 3H), 2.42 (s, 3H), 1.95 (d, *J* = 1.2 Hz, 3H).