Supporting Information

General Chemical Methods: Solvents and reagents were purchased from commercial suppliers and used without additional purification. ^{1}H and ^{13}C NMR spectra were recorded at 400 MHz and 100 MHz, respectively, in DMSO-d_6. Chemical shifts, δ, are reported in ppm and coupling constants, *J*, are expressed in Hertz (Hz). Abbreviations for peaks are s = singlet, d = doublet, t = triplet, and m = multiplet. High-resolution mass spectrometry was performed using positive mode electrospray ionization methods (ESMS) with a Bruker BioTOF II spectrometer.

Synthesis of 2-((5-chloro-2-((2-methoxy-4-(1-methylpiperidin-4-yl)phenyl)amino)pyrimidin-4-yl)amino-*N*-methylbenzenesulfonamide (compound 19): Commercially available 2-((2-amino-5-chloropyrimidin-4-yl)amino)-*N*-methylbenzenesulfonamide (CAS 761440-11-3, Chemscene) (50.8 mg, 153 umol) and commercially available 2-methoxy-4-(1-methylpiperidin-4-yl)aniline (CAS 1124330-14-8, A Chemtek) (68.6 mg, 311 umol) were dissolved in 1.0 mL of 0.625 M HCl in dry ethanol in a dry 5 mL vial with a stir bar under nitrogen. The vial was capped and stirred at 120 °C for 6 h. The mixture was poured into an aqueous potassium carbonate solution (10 mL 10% K_2CO_3) and extracted with ethyl acetate (3 x 20 mL). The organic phase was concentrated and purified by column chromatography on silica gel using dichloromethane-methanol gradient elution (dichloromethane containing 1% triethylamine, gradient 0-10% methanol) to yield **19** as a pale viscous oil, 40 mg, 77 umol, 50%, with purity of 98%. ^{1}H NMR (400 MHz, DMSO-d_6) δ 9.34 (s, 1H), 8.47 (d, *J*=8.3 Hz, 1H), 8.24 (s, 1H), 8.21 (s, 1H), 7.78 (d, *J*=7.9 Hz, 2H), 7.61 (d, *J*=8.1 Hz, 1H), 7.52 (t, *J*=7.8 Hz, 1H), 7.26 (t, *J*=7.6 Hz, 1H), 6.92 (s, 1H), 6.77 (d, *J*=8.2 Hz, 1H), 3.79 (s, 3H), 3.32 (s, 2H), 2.93 (d, *J*=11.1 Hz, 2H), 2.43 (s, 3H), 2.25 (s, 3H), 2.06 (t, *J*=10.1 Hz, 2H), 1.80-1.65 (m, 3H). ^{13}C NMR (100 MHz, DMSO-d_6) δ 158.2, 155.0, 154.9, 150.9, 142.5, 136.3, 133.1, 128.9, 126.6, 126.0, 123.2, 123.0, 122.9, 117.9, 109.8, 104.7, 55.7, 55.5, 45.9, 40.9, 32.8, 28.5. HRMS: calcd for $C_{24}H_{29}ClN_6NaO_3S$ $[M + Na]^+$, 539.1608; found 539.1608.

Compound purity: The purity of all compounds in **Tables 1-4** was determined by analytical reverse-phase high performance liquid chromatography (HPLC) analysis using a Waters 2695 HPLC (column: Phenomenex Synergi Fusion-RP, 4.0 µm, 4.6 mm × 150 mm); mobile phase gradient starting at 50:50 0.1% TFA in H_2O:MeOH, gradient to 10:90 0.1% TFA in H_2O:MeOH over 20 minutes, hold at 10:90 0.1% TFA in H_2O:MeOH for 4 minutes, reset to 50:50 0.1% TFA in H_2O:MeOH hold 6 minutes; detector, Waters 2996; injector, automated injector; detection wavelength, 254 nm; flow rate, 1.0 mL/min.; ambient temperature. Purity of compounds >95%.

Table S1. Inhibitory potencies of pyrrolopyrimidines at TSSK2 compared to other kinases

Compound	GSK ID #	Kinase	IC_{50}, nM	Reference
10	GSK2163632A	TSSK2	22	Current study
		IGF-1	0.2	Compound 28[10a]
		GRK1	130	[10d]
		GRK2	20,000	[10d]
		GRK5	3200	[10d]
		PKA	>500,000	[10d]
11	GSK2110236A	TSSK2	47	Current study
		IGF-1	< 0.2	Compound 27[10a]
		GRK1	630	[10d]
		GRK2	20,000	[10d]
		GRK5	3200	[10d]
		PKA	>500,000	[10d]

Compound	GSK ID #	Kinase	IC_{50}, nM	Reference
1	GSK1220512A	TSSK2	72	Current study
		IGF-1	0.8; 0.9	Cmpd 25[10c]; Compd 4[10a]
5	GSK1326255A	TSSK2	107	Current study
		IGF-1	0.3	Compound 9[10c]
		GRK1	500,000	[10d]
		GRK2	7900	[10d]
		GRK5	2500	[10d]
		PKA	>500,000	[10d]
2	Compound 6[10a]	TSSK2	150	Current study
		IGF-1	1.3	Cmpd 26[10c]; Cmpd 6[10a]
12	GSK2220400A	TSSK2	247	Current study
		IGF-1	1.3 - 4	Example 84[10f]
		Insulin R	0.25-1	Example 84[10f]
		ALK	1.3 - 4	Example 84[10f]
		GRK1	10,000	[10d]
		GRK2	200,000	[10d]
		GRK5	6300	[10d]
6	GSK1173862A	TSSK2	250	Current study
		IGF-1	0.4	Compound 8[10c]
7	GSK2213727A	TSSK2	280	Current study
		IGF-1	0.5	Compound 11[10b]
		JNK1	1995	Compound 11[10b]
3	GSK1392956A	TSSK2	1200	Current study
		IGF-1	16	Compound 32[10c]
13[a]	GSK1838705A	TSSK2	1300	Current study
		IGF-1	2	[10e]
		Insulin R	1.6	[10e]
		ALK	0.5	[10e]
		CLK2	21	[10e]
		Fes	16	[10e]
		MLCK	34	[10e]
		IRR	49	[10e]
		Fer	87	[10e]
		FAK	195	[10e]
		CHK2	215	[10e]
		TSSK2	452	[10e]
9	GSK1713088A	TSSK2	1300	Current study
		IGF-1	25	Compound 9[10b]
		JNK1	3981	Compound 9[10b]
		GRK1	13,000	[10d]
		GRK2	6300	[10d]
		GRK5	2500	[10d]
		PKA	>500,000	[10d]
15	GSK1511931A	TSSK2	1500	Current study
		IGF-1	13	Compound 13[10a]
14	GSK2186269A	TSSK2	4800	Current study
		IGF-1	0.8	Compound 33[10a]
16	GSK1751853A	TSSK2	6000	Current study
		IGF-1	32	Compound 31[10b]

Compound	GSK ID #	Kinase	IC_{50}, nM	Reference
8	GSK2219385A	JNK1	1585	Compound 31[10b]
		TSSK2	14,000	Current study
		IGF-1	63	Compound 13[10b]
		JNK1	10,000	Compound 13[10b]

[a] K_D values for compound **13** (GSK1838705A) binding to 78 kinases have been reported, with affinities ranging from 0.55 nM for ALK to 8500 nM for IRAK1.[11]

Table S2. Inhibitory potencies of pyrimidines at TSSK2 compared to other kinases

Compound	Name	Kinase	IC_{50}, nM	Reference
17	ALK inhibitor 1	TSSK2	31	Current study
		FAK	2	Example 3-39[13p]
		IGF-1R	90	Example 3-39[13p]
18	ALK inhibitor 2	TSSK2	37	Current study
		FAK	5	Example 20-06[13p]
35	Mps1-IN-3	TSSK2	58	Current study
		MPS1	50	[13a]
		LTK (TYK1)	18	[a]
		IGF-1R	26	[a]
		INSRR (IRR)	29	[a]
		CLK2	42	[a]
		PTK2 (FAK)	74	[a]
		EGFR (ErbB1)	289	[a]
		CLK1	443	[a]
		AURKB (Aurora B)	2100	[a]
22[b]	TAE684	TSSK2	80	Current study
		ALK	3.7	[12]
		FLT3	3; 182	[12, 13b]
		LRRK2	7.8	[13c]
		Tie2	12	[13b]
		InsR	43.7; ~10-20	[12, 13b]
		FGFR-3K650E	41	[12]
		KDR	89	[12]
		cFes	118	[13e]
		FAK	270	[12]
		Tek	219	[12]
		Syk	286	[13b]
		TRKB	422	[13b]
		CDK1/B	490	[12]
		Bmx	600	[13b]
24	ALK-IN-1	TSSK2	230	Current study
		ALK	0.07	Compound 11L[13f]
		IGF-1R	3.2	Compound 11L[13f]
		InsR	100	Compound 11L[13f]
25	Brigatinib; AP26113	TSSK2	510	Current study
		ALK	0.37	Compound 11Q[13f]
		IGF-1R	24.9	Compound 11Q[13f]

Compound	Name	Kinase	IC_{50}, nM	Reference
		IRE1a	66	[13g]
		InsR	196	Compound 11Q[13f]
23	Ceritinib; LDK378	TSSK2	610	Current study
		ALK	26	Compound 15b[12]
		InsR	319.5	Compound 15b[12]
34	AZD3463	TSSK2	690	Current study
		IRE1a	710	[13g]
20	CZC-54252	TSSK2	750	Current study
		LRRK2	1.28	[13h]
26	CTx-0294885	TSSK2	963	Current study
		Flt3	1	[13d]
		Src	2	[13d]
		JAK2	3	[13d]
		VEGFR3	3	[13d]
		FAK	4	[13d]
36	ASP3026	TSSK2	3800	Current study
		ALK	3.5	[13i]
30	TG101209	TSSK2	6200	Current study
		JAK-2	6	[13j]
		BRD4	130	[13k]
27	TAE 226	TSSK2	7600	Current study
		FAK	17, 7, 5.5	[13l, 13p, 14]
		Flt3	26	[13d]
		IGF-1R	~140	[13l]
		EGFR	326	[13m]
21	CZC-25146	TSSK2	13,000	Current study
		LRRK2	4.76	[13h]
31	Rociletinib; Co 1686	TSSK2	15,000	Current study
		EGFR	91[c]	[13n]
32	KRCA-0008	TSSK2	62,000	Current study
		ALK	3.9	[13o]
28	GSK1576028A; CTx-0152960	TSSK2	>100,000	Current study
		Flt3	1	[13d]
		Src	2	[13d]
		JAK2	2	[13d]
		FAK	4	[13d]
		VEGFR3	5	[13d]

[a] Personal communication, Jinhua Wang and Nathaniel Gray, Harvard University.

[b] K_D values **22** for binding to 343 kinases have been reported, with affinities ranging from 0.49 to 9700 nM.[11]

[c] K_D value.

Table S3. Compound **19** kinase profiling. Compound **19** (100 nM) was tested in a broad panel of 369 kinases for substrate phosphorylation assays in duplicate using 10 μM [γ^{33}P]ATP at Reaction Biology Corp. Kinases inhibited to a greater extent than TSSK2 are listed

Kinase	% inhibition (100 nM)
GLK/MAP4K3	101
ALK	100
FER	100
FES/FPS	100
IR	100
IRR/INSRR	100
LOK/STK10	99
FAK/PTK2	99
IGF1R	99
TNK1	99
STK22D/TSSK1	99
LRRK2	98
FLT3	98
ROS/ROS1	98
RSK2	98
CAMK2a	98
HPK1/MAP4K1	97
TYK1/LTK	97
ARK5/NUAK1	97
MYO3b	96
RSK3	96
ULK1	95
PYK2	95
CAMK2d	95
SIK2	94
RSK4	93
ULK2	90
PHKg1	90
DDR1	90
FLT4/VEGFR3	90
RSK1	89
MARK4	88
CHK2	88
FMS	88
SNARK/NUAK2	87
ACK1	87
TAOK1	86
SIK1	86
CAMK2b	86
CDK5/p35	85
CLK1	85
MARK2/PAR-1Ba	84
MLK1/MAP3K9	83
PDGFRb	83
CDK5/p25	83

Kinase	% inhibition (100 nM)
TYK2	82
KHS/MAP4K5	82
AXL	81
FGFR2	81
PKCnu/PRKD3	80
FRK/PTK5	80
MARK1	80
PHKg2	79
YES/YES1	79
MARK3	79
JAK3	79
CDK19/cyclin C	79
NEK9	77
BRK	77
PKD2/PRKD2	76
SYK	75
PDGFRa	75
TSSK2	75

Table S4. Compound **19** inhibition of off-target kinases and TSSKs. Compound **19** was tested at 10 concentrations using substrate phosphorylation assays in singlicate using 10 μM [γ^{33}P]ATP at Reaction Biology Corp. Staurosporine was used a reference compound

Kinase	Compound 19 IC_{50}, nM	Staurosporine IC_{50}, nM
ALK	0.63	1.7
FES	0.36	2.4
IRR/INSRR	0.68	15
FAK	6.6	15
TSSK1	0.86	0.036
TSSK2	40	3.4
TSSK3	700	27
TSSK6	84,000	320