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Discovery of IACS-9439, a potent, exquisitely selective and orally bioavailable inhibitor of CSF1R

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Keywords: CSF1, CSF1R, CSF1R inhibitor, colony-stimulating factor 1, immune-oncology, macrophage, tumor associated macrophage, TAM

ABSTRACT

Tumor-associated macrophages (TAMs) have a significant presence in the tumor stroma across multiple human malignancies and are believed to be beneficial to tumor growth. Targeting CSF1R has been proposed as a potential therapy to reduce TAMs, especially the pro-tumor, immune-suppressive M2 TAMs. Additionally, high expression of CSF1R on tumor cells has been associated with poor survival in certain cancers, suggesting tumor dependency and therefore a potential therapeutic target. The CSF1–CSF1R signaling pathway modulates the production, differentiation and function of TAMs; however, the discovery of selective CSF1R inhibitors devoid of Type III kinase activity has proven to be challenging. We discovered a potent, highly selective and orally bioavailable CSF1R inhibitor, IACS-9439 (**1**). Treatment with **1** led to a dose dependent

reduction in macrophages, promoted macrophage polarization toward the M1 phenotype, and led to tumor growth inhibition in MC38 and PANC02 syngeneic tumor models.

INTRODUCTION

Strategies to enhance anti-tumor immunity by reactivating the adaptive and innate immune compartments through checkpoint inhibitors against programmed cell death 1 (PD1), PD1 ligand (PDL1) and cytotoxic T lymphocyte antigen 4 (CTLA4) have shown favorable clinical responses, yet, only a fraction of patients show durable responses. While tumor-intrinsic resistance mechanisms may exist, clinical and preclinical evidences highlight the abundance of tumor-associated macrophages (TAMs) as critical regulatory immune cells that promote tumor progression. Macrophages exist primarily in two main polarization states with the alternatively activated M2 TAM responsible for promoting tumor progression by secreting anti-inflammatory cytokines such as IL-10 and TGFβ, whereas the classically activated M1 promote immune-mediated tumor killing through the production of pro-inflammatory cytokines such as IL-1β, IL-6 and tumor necrosis factor α (TNF-α).[1-3] To overcome the immunosuppressive and pro-tumoral functions of TAMs,

therapeutic strategies have focused on TAM depletion in the tumor microenvironment as well as TAM reprogramming to favor anti-tumoral functions (polarization from M2 to M1).[4, 5]

Colony-stimulating factor 1 receptor (also known as CSF1R, or macrophage colony-stimulating factor receptor, M-CSFR) is a membrane-associated tyrosine kinase. It acts as the receptor for colony-stimulating factor 1 (CSF1), a cytokine which controls the production, differentiation and function of macrophages. Ligand binding activates CSF1R through a process of oligomerization within the membrane, transphosphorylation of the intracellular domain, and subsequent signaling.[6]

In several preclinical studies using murine breast cancer and glioblastoma models, it was demonstrated that CSF1–CSF1R signaling blockade slowed primary tumor growth, reduced metastatic potential and improved the long-term survival of tumor-bearing mice.[7, 8,] Treatment with BLZ945, a potent CSF1R inhibitor, attenuated tumor growth, correlated with decreased TAM presence, and enriched CD8+ T cells in tumor stroma in a murine K14-HPV-16 transgenic mouse model of cervical carcinogenesis.[9] These findings have been translated into a first-in-human Phase I/II study of BLZ945 alone or in combination

with PDR001, a monoclonal antibody against PD-1 in advanced solid tumors (NCT02829723).[10,11] Pexidartinib (PLX3397), an unselective CSF1R inhibitor that also inhibits cKIT, FLT3, PDGFRα and PDGFRβ with low nanomolar activity, recently received FDA approval.[12] Pexidarinib demonstrated reduction of TAMs, efficient target engagement, and clinical efficacy as a monotherapy, with a 38% overall response rate in pigmented villonodular synovitis (PVNS), an orphan disease characterized by overexpression of CSF1R.[12-16] Additionally, emactuzumab (RG7155), a neutralizing antibody against CSF1R not only reduced macrophage infiltration in mouse models but also demonstrated similar therapeutic effects against diffuse-type giant cell tumors in patients.[17] Two other neutralizing antibodies against CSF1R, AMG 820 (NCT01444404), and IMC-CS4 (NCT01346358) are also currently in Phase I clinical trials for the treatment of advanced solid tumors.[18, 19] While there are several clinical opportunities for single agent activity by suppressing CSF1R biology in dependent tumor cells, full clinical benefit is likely to come from combining CSF1R inhibition in macrophages with immune modulating agents targeting T-cell biology, such as anti-CTLA4, anti-PD-1 and anti-PD-L1 therapeutics. These combinations have the potential to promote tumor suppression

through modulation of multiple arms of the immune system. These combinations currently are being explored in numerous clinical trials (i.e. NCT02777710, NCT04301778 and NCT03927105).[20-22]

Several small molecule CSF1R inhibitors have been described in the literature, however, only a few were reported to possess high levels of selectivity (Figure 1).[7-16, 23-31] BLZ945 was shown to inhibit CSF1R enzymatic activity with an IC_{50} = 1 nM, and was reported to have a >3200-fold selectivity for CSF1R over other related kinases.[8] GW2580 inhibited CSF1R enzymatic activity with an IC_{50} = 30 nM, showing 22-fold and 75-fold selectivity towards TrkB and C, respectively, and over 150-fold selectivity against other kinases.[29,32] GW2580 displayed an unfavorable in vivo PK profile with low exposures and rapid clearance, and has not been further developed.[29] A structurally related, highly selective inhibitor with an azetidine scaffold, JTE-952 was also reported in the literature. In a panel consisting of 51 kinases, JTE-952 was reported to have a 20-fold selectivity for CSF1R over TrkA, however, selectivity for TrkB and TrkC was not reported.[30, 31] Analysis of the crystal structure of PLX3397 bound to CSF1R (PDB 4R7H)[15] and protein-ligand binding models of other CSF1R inhibitors (using PDB 3LCO) suggested that these

inhibitors bind in the ATP binding pocket in the inactive, DFG-out conformation of the kinase.

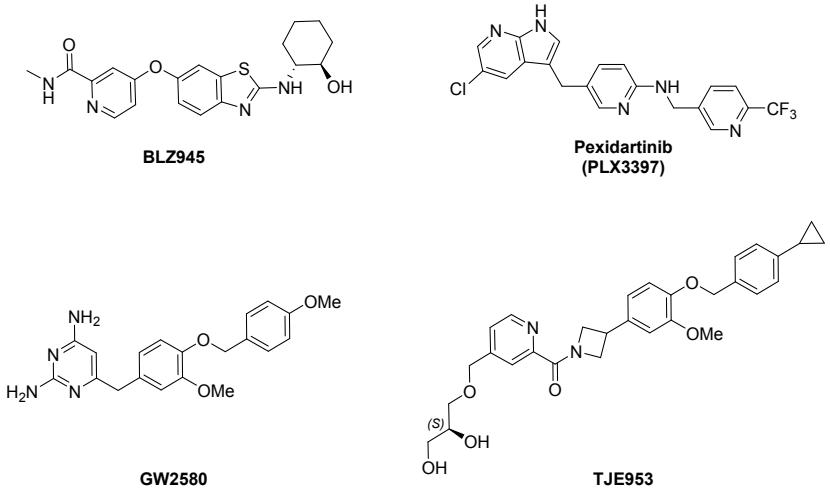


Figure 1. Representative examples of small molecule CSF1R inhibitors.

RESULTS AND DISCUSSION

The objective of our program was to discover a highly selective small molecule CSF1R inhibitor with excellent physicochemical and pharmacokinetic properties that potentially could be used in combination with other immuno-oncology therapies. Herein, we describe our efforts to identify a potent, orally bioavailable tool compound that we transformed into a highly selective and efficacious lead candidate.

At the outset of our program, we performed a scaffold hopping exercise using benzothiazole as a template, with the search directed towards finding novel combinations of hinge binding and linker groups (Figure 2A); BREED[33], as implemented in MOE[34], was used for this purpose. A set of 1,945 pre-aligned kinase-ligand complexes from the PDB were compared with the benzothiazole in a molecular model of BLZ945 bound to CSF1R, resulting in novel compounds that maintained both the position of the benzothiazole core and the interactions with the kinase hinge region. Examination of these results suggested that the kinase hinge residue, Cys666, could be engaged efficiently using two-atom linkers placed between the benzothiazole core and suitable hinge-binding moieties (Figure 2B).

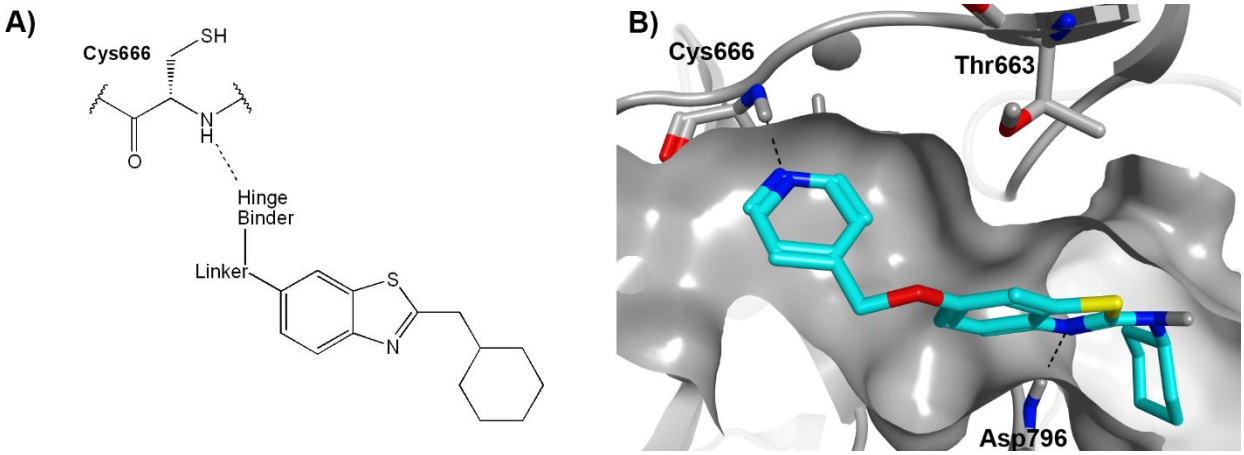
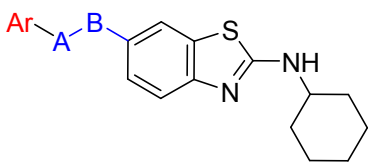
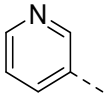
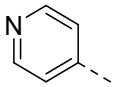


Figure 2. A) Schematic showing of scaffold hopping template and kinase hinge target interactions B) Molecular model of 4-pyridyl hinge binding motif with -C-O- linker engaging the backbone NH of Cys666 (PDB 3LCO).

To test this hypothesis, a focused SAR study utilizing a variety of two-atom linkers was performed. This study revealed that the -C-O- linker was preferred when combined with a 4-pyridyl hinge binding motif resulting in compound **3**, which exhibited an IC_{50} of 303 nM in a CSF1R FRET-based displacement binding assay (Table 1). There was less preference among the two-atom linkers when combined with the 3-pyridyl hinge-binding moiety; none of these compounds matched the enzymatic activity of compound **3**.

Table 1. Enzymatic activity of compounds containing a two-atom linker between the hinge-binding pyridine and the benzothiazole core.



A-B \ Ar		
-C-O- CSF1R IC_{50} (nM)	**2** 781	**3** 303
-O-C- CSF1R IC_{50} (nM)	**4** 450	**5** 4,413
-NH-C- CSF1R IC_{50} (nM)	**6** 655	**7** 27,712

In an effort to improve potency, an amine substituent was incorporated onto the pyridine of compound **3**, which was predicted to form a second hinge interaction with the backbone carbonyl of Cys666. Indeed, this modification led to a six-fold improvement in enzymatic potency as demonstrated by compound **8**. Incorporating a chloro group at the three position of the pyridine led to similar improvement in potency as shown by compound **9**. Combining these two substituents had an additive effect with a 75-fold improvement in potency relative to compound **3**, with compound **10** exhibiting an IC_{50} of 4 nM in the CSF1R FRET-based displacement binding assay. Compound **10** also inhibited mCSF1-

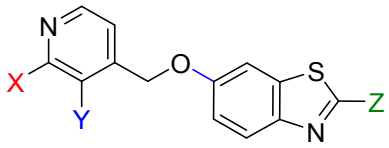
mediated proliferation of MNFS-60 myelogenous leukemia cells with an IC_{50} of 228 nM, while it did not show significant activity against mCSF1-independent NS0 cell proliferation (Table 2).

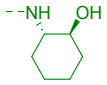
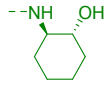
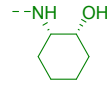
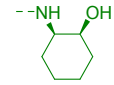
Our molecular models predicted that the cyclohexyl ring extended into the deep pocket of the DFG-out conformation of CSF1R, a region that provided additional opportunities to improve potency and selectivity, given the modest differences in sequence among type III kinases in this area (ie. Met637 is a Leu in cKIT, G795 is a Cys in other type III kinases). To engage the side chain of Glu633 in the deep pocket region, a hydroxyl substituent was incorporated on the cyclohexyl ring. To maximize the probability of engaging this interaction, we examined all four stereoisomers of the 2-amino cyclohexan-1-ol group. However, incorporating the hydroxyl group did not make a significant contribution to enzymatic or cellular potency.

To evaluate whether incorporation of the hydroxyl group on the cyclohexane ring offered any selectivity advantage, and whether the orientation of the hydroxyl group affected selectivity, we evaluated these inhibitors in a binding assay against a representative set of type III kinases that are related to CSF1R, including FLT3, cKIT, PDGFRα and

PDGFRβ. Overall, compound **12** exhibited a favorable selectivity profile. The cellular activity of compound **12** was measured by monitoring inhibition of CSF1R autophosphorylation after CSF1 stimulation of THP-1 cells, a human monocytic cell line. In order to assess the compound's selectivity against PDGFRβ, we measured agonist (PDGF-DD) induced PDGFRβ autophosphorylation in a HEK293 cell line expressing human recombinant PDGFRβ (HEK293/PDGFRβ cells). BLZ945 was also tested in this assay as a competitor with reported selectivity against PDGFRβ. Our results confirmed that both BLZ945 and **12** exhibited minimal selectivity against PDGFRβ in a cellular setting, with **12** inhibiting CSF1 stimulated CSF1R autophosphorylation in THP-1 cells with an IC_{50} of 183 nM, while inhibiting PDGFRβ autophosphorylation in HEK293/PDGFRβ cells with an IC_{50} of 446 nM. BLZ945 displayed an IC_{50} of 155 nM in the cellular phospho-CSF1R (THP) assay and IC_{50} of 579 nM in the phospho-PDGFRβ (HEK293/ PDGFRβ) assay.

Table 2. SAR studies in the hinge region and deep pocket area of the 6-(pyridin-4-ylmethoxy)benzo[d]thiazole scaffold lead to significant improvement in potency.



Compound	8	9	10	11	12	13	14
X	$-NH_2$	-H	$-NH_2$	$-NH_2$	$-NH_2$	$-NH_2$	$-NH_2$
Y	-H	-Cl	-Cl	-Cl	-Cl	-Cl	-Cl
Z							
CSF1R IC_{50} [nM]	52	30	4	2	1.4	1.1	2.4
CSF1R Kd [nM] (fold)	ND	ND	2.5	1.1	0.6	1.2	1.9
cKIT Kd [nM] (fold)	ND	ND	79 (31)	44 (40)	180 (300)	170 (141)	1000 (526)
FLT3 Kd [nM] (fold)	ND	ND	170 (68)	65 (59)	74 (86)	400 (333)	320 (168)
PDGFRα Kd [nM] (fold)	ND	ND	46 (18)	1.3 (1.2)	12 (20)	28 (23)	94 (49)
PDGFRβ Kd [nM] (fold)	ND	ND	3.8 (1.5)	0.84 (0.76)	19 (32)	1.2 (1)	5.6 (3)
Phospho-CSF1R (THP-1) IC_{50} [nM]	ND	ND	ND	ND	183	ND	ND

Phospho-PDGFRβ (HEK293) IC_{50} [nM] (Fold X)	ND	ND	ND	ND	446 (2.4)	ND	ND
MNSF60 IC_{50} [nM]	2269	1204	228	50	133	98	189
NS0 IC_{50} [nM]	18,809	14,989	41,666	27,500	47,945	ND	ND

To assess broad kinase selectivity, compound **12** was evaluated against a panel of 468 kinases (Figure 3). It exhibited an acceptable selectivity profile, with significant activity against only a handful of related type III kinases at 1 uM including c-KIT, FLT3, PDGFRα and PDGFRβ (Table S1).

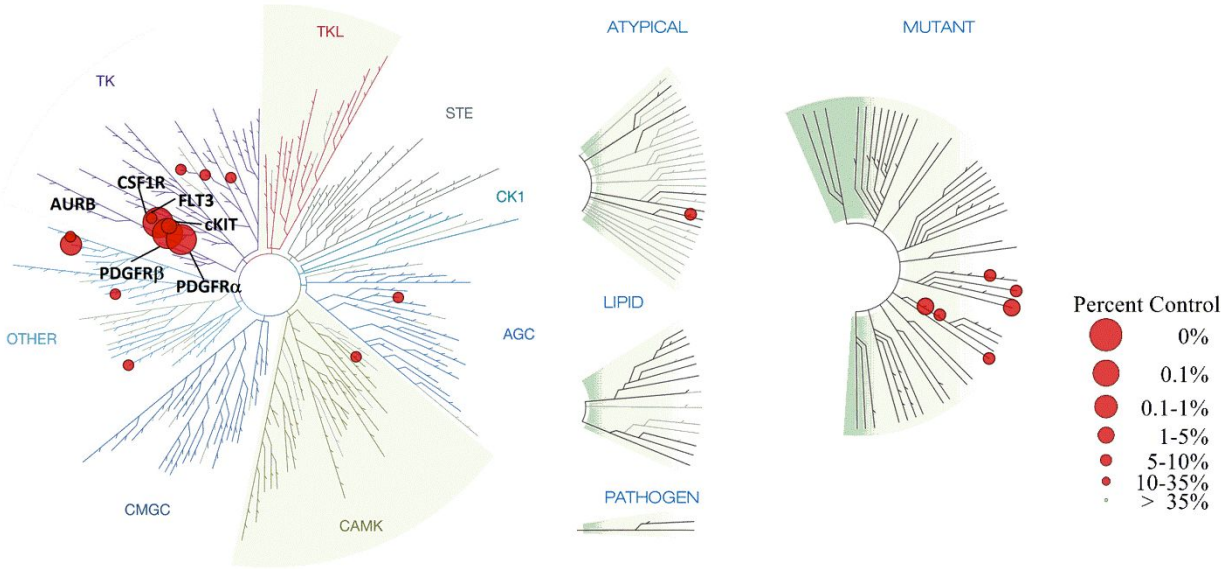


Figure 3. Selectivity profile of compound **12** against a panel of 468 kinases at 1 μM test concentration. The larger the red circle, the greater the inhibition. Six kinases with percent of control < 10: CSF1R, cKIT, FLT3, AURB, PDGFRα and PDGFRβ. See also Table S1, Supporting information.

Compound **12** displayed low to moderate microsomal and hepatic clearance in *in vitro* microsomal and hepatocyte preparations and high plasma protein binding. It exhibited a suitable *in vivo* PK profile across species, with low clearance, reasonable half-life and good oral bioavailability (Table 3).

Table 3. Microsomal and hepatic stablity, plasma protein binding and *in vivo* PK profile of compound **12**.

Specie	*In vitro* ADME properties of **12**	*In vivo* pharmacokinetic data of compound **12**

s	Plasma protein binding (% bound)	Hepatocytes CLint,scaled [mL/min	Microsomes CLint,scaled [mL/min	Dose IV/PO [mg/kg]	CL [mL/min/kg] (IV)	Vss [L/kg]	$T_{1/2}$ [h] (IV)	Oral F [%]
Mouse	>99	11	55	0.3/10	23	1.8	1.6	>90
Rat	>99	30	68	0.3/3	3.2	0.75	3	67
Dog	>99	155	293	0.3/3	12	1.3	4.8	34
Monke	>99	58	95	0.3/1	1.2	0.6	7.0	72
Human	>99	10	25					

Given its *in vitro* and *in vivo* profiles, compound **12** was used to probe CSF1R biology, with BLZ945 serving as a comparitor in these experiements. An immunohistochemistry (IHC) assay was used to measure the well-established macrophage marker F4/80 in PANC02 tumors from mice that had been treated with daily oral doses of 200 mg/kg compound **12** or BLZ945 for 7 days (n=4 mice/group). Treatment with each compound resulted in similar extent of reduction of F4/80 positive cells within the tumors (Figures 4A-D), consistent with macrophage depletion. As an orthogonal measure of compound activity, CSF1R protein levels were measured by ELISA in tumors from mice similarly treated, and were found to be depleted to a similar extent by both compounds (Figure 4E) (n=4 mice/group). Furthermore, as an additional measure of anti-macrophage activity, mRNA transcripts of well known macrophage expressed genes were measured, and

similar to the IHC and ELISA results, 200 mg/kg of compound **12** or BLZ945 (n=3 mice/group) reduced the expression of the panel by 62-93% (Figure 4F). Similar depletion of macrophages was observed for MC38 and EMT6 tumors treated with compound **12** or BLZ945 (data not shown). Taken together, treatment of PANC02 tumors with compound **12** reduced tumor macrophages to a similar extent as BLZ945.

Treatment of mice bearing MC38 allograft tumors with 200 mg/kg compoud **12** (n=10 mice/group) resulted in 62% reduction in tumor growth (Figure 4G), consistent with the notion that inhibition of CSF1R has therapeutic potential, possibly through macrophage depletion.

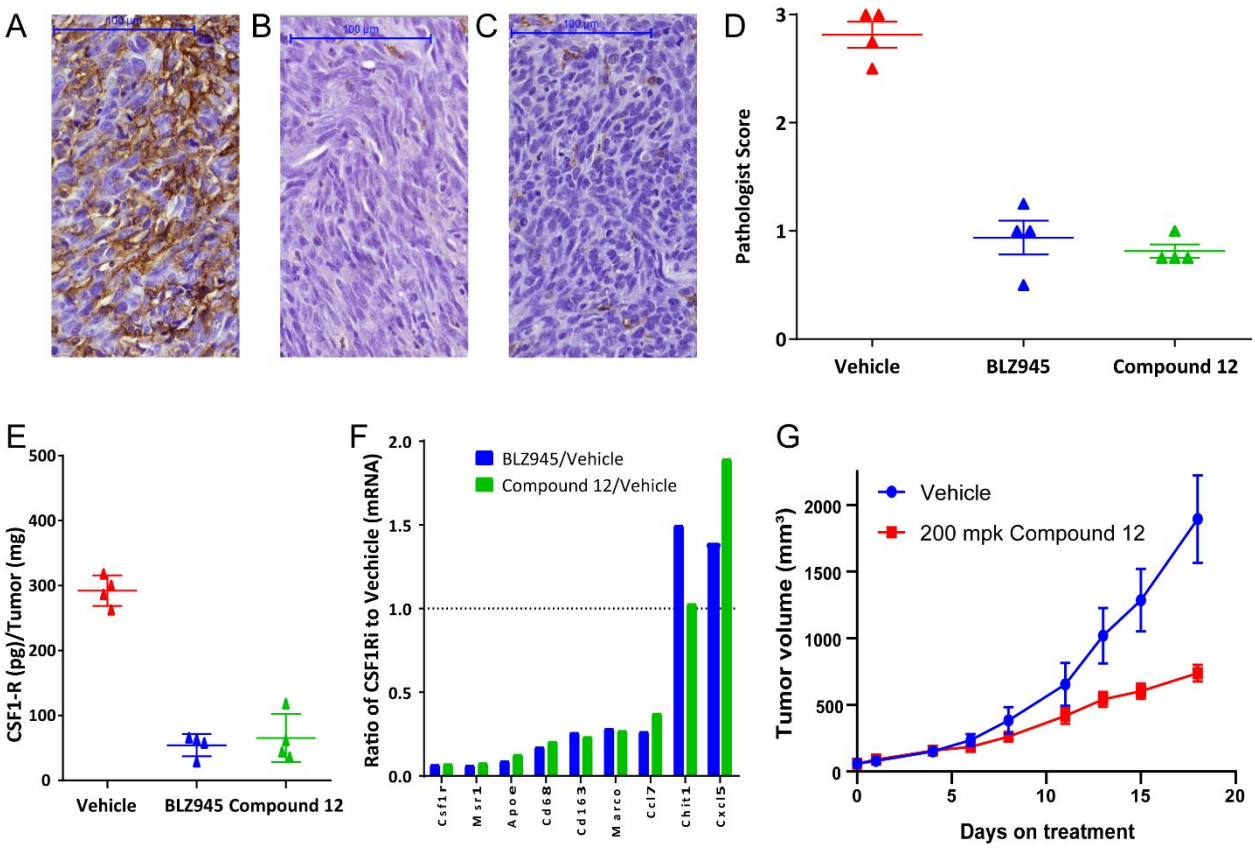


Figure 4. CSF1R inhibitors depletes tumor macrophages and reduces tumor growth. IHC for F4/80 for PANC02 tumors in mice that were treated daily for 7 days with (A) vehicle, (B) 200 mg/kg BLZ945 or (C) 200 mg/kg compound **12** (n=4 mice/group). (D) Pathologist scored quantification of F4/80 IHC signal in PANC02 tumors from mice treated daily for 7 days with vehicle, 200 mg/kg BLZ945 or 200 mg/kg compound **12** (n=4 mice/group). (E) Relative CSF1R protein expression in PANC02 tumors from mice treated daily for 7 days with vehicle, 200 mg/kg BLZ945, or 200 mg/kg compound **12** as measured using ELISA

(n = 4 mice/group). (F) Relative mRNA expression of known macrophage genes in PANC02 tumors from mice treated daily for 7 days with vehicle, 200 mg/kg BLZ945, or 200 mg/kg compound **12** (n=3 mice/group) as measured using a nanostring code set. (G) Growth of MC38 tumors implanted subcutaneously in mice and treated daily with vehicle or 200 mg/kg compound **12** (n=10 mice/group).

While the anti-tumor impact of compound **12** was encouraging, we were not satisfied with the overall potency, physicochemical properties and the level of selectivity that this inhibitor exhibited toward related type III RTKs, especially toward PDGFRβ. Therefore, we continued our quest to identify inhibitors with significantly improved potency, properties and selectivity. In order to improve the selectivity profile of the benzothiazole scaffold, we used a combination of X-ray structures from the PDB and the patent literature. Sequence and structural comparison of the CSF1R active site with c-KIT, FLT3, PDGFRα and PDGFRβ[35] revealed that CSF1R is unique among these kinases; the CSF1R Gly795 residue aligns with a cysteine in the other related kinases (Figures 5A and 5B). Substitution at the 4-position of the benzothiazole core would fill the space

adjacent to Gly795 in CSF1R, while this substitution pattern would not be tolerated in c-KIT, FLT3, PDGFRα or PDGFβ as it would lead to a steric clash with the cysteine residue (Figure 5C). Furthermore, overlaying models of compound **12** and GW2580, (Figure 5D), an inhibitor that showed high selectivity toward CSF1R over other related kinases, but not TrkA, led to the same conclusion.

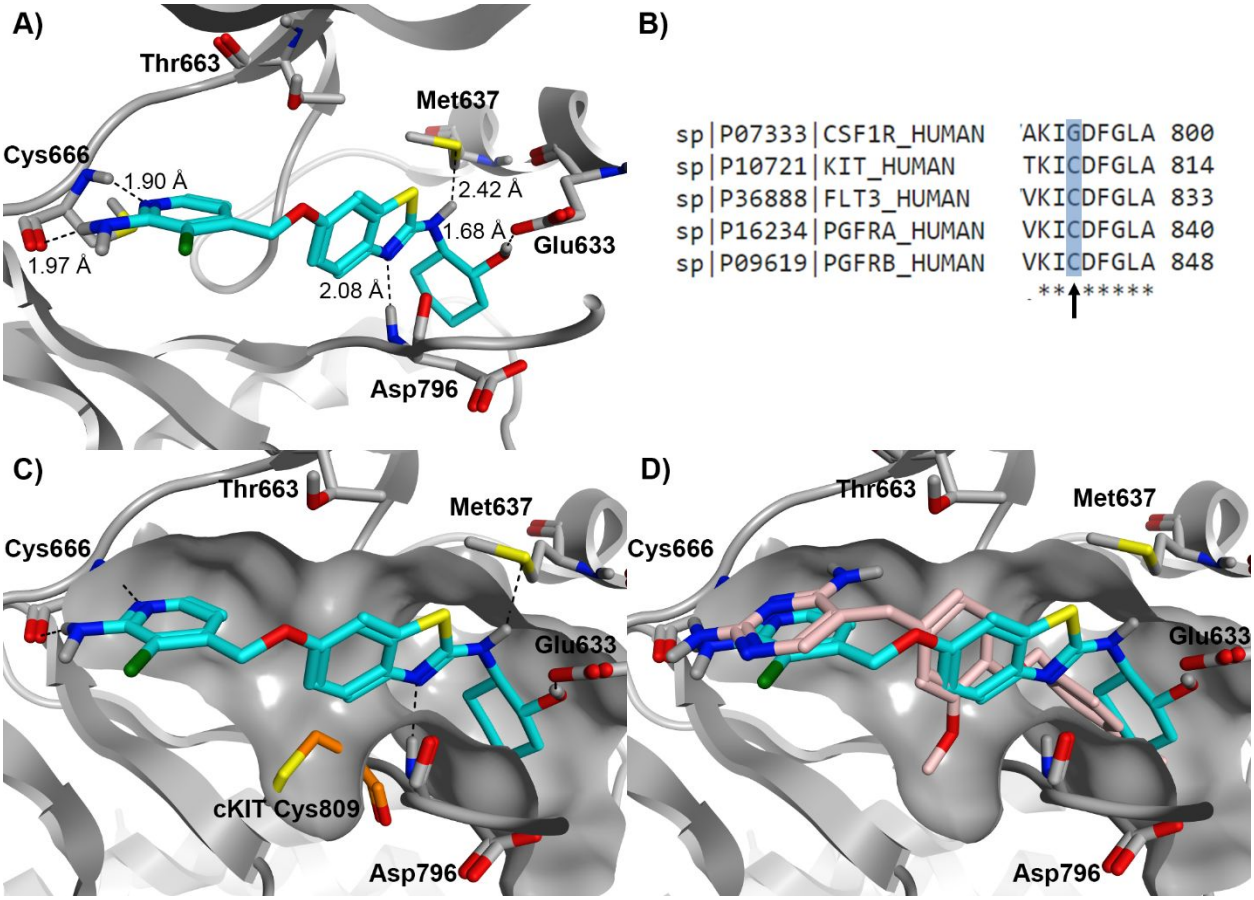


Figure 5. A) Model of compound **12** bound to CSF1R (PDB 3LCO). B) Sequence alignment of class III receptor tyrosine kinases highlighting a key difference in the xDFG

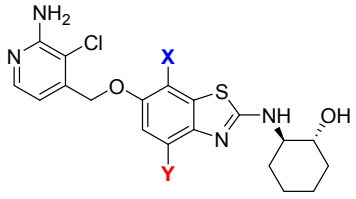
motif. C) Overlay of cKIT Cys809 (orange; PDB 4HVS) with model of compound **12** bound to CSF1R shows a small pocket that is present in CSF1R that is filled by a cysteine side chain in other class III RTKs. D) Overlay of GW2580 (pink) with model of compound **12** shows a methoxy group filling this pocket.

To test this hypothesis, we synthesized the 4-methoxy substituted derivative of **12**, compound **15**. Indeed, this modification maintained high affinity towards CSF1R in binding and cellular activity assays, and led to a dramatic improvement in selectivity against related kinases cKIT, FLT3, PDGFRα and PDGFRβ. The same high level of selectivity was observed in the cellular phospho-PDGFRβ target engagement assay (Table 4).

To understand whether introducing other substituents into the 4-position would lead to the same level of selectivity increase, we performed a focused SAR study. We found that introduction of a trifluoromethoxy substituent into the 4-position (**16**) leads to decreased cellular potency. Incorporation of a fluoro- (**17**) or chloro (**18**) substituent was tolerated for potency, however, only lower levels of selectivity was achieved compared to that of

the methoxy-substitued analog **15**. Further structural analysis revealed that a hydrophobic pocket formed by the sidechains of Thr663 and Met637 could potentially accommodate a substituent in the 7-position of the benzothiazole core and that filling this pocket could lead to a potency improvement. To test this hypothesis, the 7-chloro- (**19**) and 7-fluoro benzothiazole (**20**) analogs were prepared. These inhibitors revealed that while a substituent in the 7-position was indeed tolerated for potency, this modification was detrimental to selectivity.

Table 4. SAR study of substituted benzothiazole analogs leads to dramatic improvement of selectivity.



Compound #	15	16	17	18	19	20
X	-H	-H	-H	-H	-Cl	-F
Y	-OMe	$-OCF_3$	-Cl	-F	-H	-H

CSF1R IC50 [nM]	4	17	1	1	1.2	1.1
CSF1R Kd [nM] (Fold X)	0.6	ND	8.1	2.5	3.4	1.3
FLT3 Kd [nM] (Fold X)	30,000 (>1875)	ND	590 (72)	1300 (520)	5.4 (1.58)	23 (17)
cKit Kd [nM] (Fold X)	30,000 (>1875)	ND	>30,000 (>3703)	2100 (840)	530 (98)	790 (607)
PDGFRα Kd [nM] (Fold X)	30,000 (>1875)	ND	>30,000 (>3703)	370 (148)	12 (3.5)	32 (24)
PDGFRβ Kd [nM] (Fold X)	30,000 (>1875)	ND	>30,000 (>3703)	230 (92)	4.1 (1.2)	6.7 (5.1)
MNSF60 IC50 [nM]	345	930	288	125	104	194

Phospho-CSF1R (THP-1) [nM]	243	ND	191	127	176	ND
Phospho-PDGFRβ (HEK293) [nM] (Fold X)	>50,000 (>200)	ND	4114 (20)	1339 (10)	250 (1.4)	ND
cLogP	3.5	5.2	4.32	3.8	4.3	3.8
PPB [% bound], (r/h)	>99 / 98.7	ND	>99 / >99	>99 / 98.6	ND	ND
CLint,scaled [mL/min/kg] (r/h)	99/38	ND	78/32	91/19	ND	ND

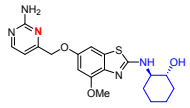
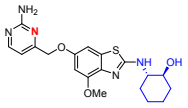
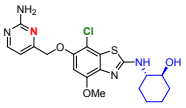
Introducing a substituent into the 4-position led to the desired selectivity improvement, however, these inhibitors possessed suboptimal *in vitro* DMPK properties, particularly unfavorable metabolic turnover in rat and human microsomes (Table 4).

Analyzing the correlation between liver microsomal intrinsic clearance and cLogP for nearly one hundred analogs that were synthesized in the 6-methoxy benzothiazole series revealed that inhibitors with a cLogP > 3.5 tended to have high clearance in human liver microsomes (Figure S1). Based on this observation we targeted the design and synthesis of inhibitors with a cLogP of less than 3.5 to improve metabolic stability, while retaining favorable enzymatic and cellular potency.

To increase polarity and modulate physicochemical properties, we devised SAR studies in the hinge as well as deep pocket area. Decreasing the cLogP by structural modifications in the hinge region quickly led to improved microsomal stability, while selectivity was preserved as demonstrated by compound **21** (Table 5). Reversing the stereochemistry of the amino- and hydroxyl- substituents on the cyclohexyl ring led to improved potency, while maintaining selectivity and favorable *in vitro* DMPK properties as shown by compound **22**. Introduction of a 7-chloro substituent onto the benzothiazole core led to further improvement in potency, as exemplified by compound **23**, without a significant deleterious effect on selectivity or physicochemical properties.

Since compounds **22** and **23** exhibited acceptable enzymatic and cellular potency, as well as a promising *in vitro* ADME profile, we decided to evaluate these inhibitors *in vivo*. Both inhibitors had a suitable *in vivo* PK profiles across species with low to moderate clearance, reasonable half-life and excellent oral bioavailability in rat and mouse, and with moderate oral bioavailability in dog (Table S2).

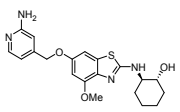
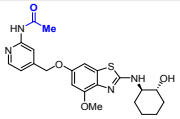
Table 5. Property optimization of benzothiazole analogs.

			
Compound #	**21**	**22**	**23**
CSF1R IC_{50} [nM]	21	7	2
CSF1R Kd [nM]	6.7	4.1	2.6
FLT3 [nM] (Fold X)	>30,000 (>4,477)	>30,000 (7,317)	>30,000 (>11,538)
cKit [nM] (Fold X)	4,800 (750)	>30,000 (7,317)	>30,000 (>11,538)
PDGFRα [nM]	4,100	>30,000	24,000

(Fold X)	(611)	(7,317)	(9,230)
PDGFRβ [nM]	4,900	>30,000	27,000
(Fold X)	(731)	(7,317)	(10,384)
MNSF60 IC_{50} [nM]	504	178	60
Phospho-CSF1R [nM] (THP-1)	486	215	63
Phospho-PDGFRβ [nM] (HEK293)	41000	>50000	34254
(Fold X)	(84)	(>23)	(543)
cLogP	2.41	2.41	3.02
PPB [% bound], (r/h)	90.4/77.1	92.2/77.6	98.3/97.2
$CL_{int,scaled}$ [mL/min/kg] (r/h)	24/10	20/9.5	43/33

These compounds exhibited excellent overall properties, however, we were not satisfied with the cellular potency of these inhibitors. SAR studies performed earlier on aminopyridine derivative **24** revealed that introduction of an acetamide onto the aminopyridine moiety of **24** resulted in a significant boost in potency (Table 6).

Table 6. Potency improvement by introducing an acetamide onto compound **24**.

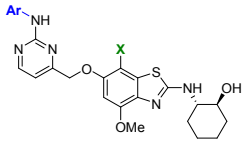
		
Compound #	**24**	**25**
CSF1R IC_{50} [nM]	76	4
MNSF60 IC_{50} [nM]	1455	82

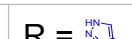
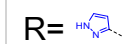
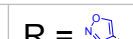
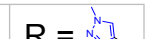
Although compound **25** possessed excellent potency, it suffered from significant non-NADPH-dependant microsomal metabolism; incubation with human and mouse liver microsomes for 45 min led to 80% and 54% parent remaining, respectively, even in the absence of NADPH (Table S4.) Metabolite identification studies in human and mouse liver microsomes confirmed that the major metabolite is the hydrolysis product of the amide of compound **25** (Table S5).

Based on this information we hypothesized that we would be able to further boost the potency, while maintaining optimal properties, if we introduced a substituent onto the amino pyrimidine moiety of inhibitor **22**. However, instead of introducing a hydrolytically unstable amide, we decided to incorporate a urea, as well as a series of heterocyclic

groups including pyrazole, triazole and pyridine. Assessing the enzymatic and cellular potency as well as microsomal stability of these inhibitors clearly indicated that the methylpyrazole analog IACS-9439 (**1**) stands out. IACS-9439 (**1**) displayed the predicted potency boost, with single-digit nanomolar potency in the MNSF-60 cell phenotypical assay and possessed reasonable metabolic stability when incubated with human and rat microsomal preparations. To test whether we could further improve the potency, we also prepared the 7-chloro analog of IACS-9439. While this analog displayed excellent potency, it had high metabolic turnover in rat and human microsomes.

Table 7. SAR studies to improve potency of selective benzothiazole analogs.



	R =  X = H	R =  X = H	R =  X = H	R =  X = H	R=  X = H	R =  X = H	R =  X = H	R =  X = H	R =  X = Cl
Compound #	**26**	**1**	**27**	**28**	**29**	**30**	**31**	**32**	**33**

CSF1R IC50 [nM]	11	1.7	9	0.81	48	2.6	5.7	626	1.2
Cell. IC50 [nM] (MNFS-60)	270	7	105	7	1304	26	48	2511	14
cLogP	2.37	3.11	3.7	2.98	3.07	3.05	3.32	3.46	3.71
CLint,scaled [mL/min/kg] (r/h)	19/23	38/14	46.2/17	120/32	ND	87/61	25/43	ND	124/65

Based on the excellent potency and selectivity profile, we decided to further evaluate IACS-9439 (**1**). To assess the selectivity, the inhibitor was tested against a panel of kinases (ScanEdge, 97-member kinase panel, DiscoverX) at 1 μM (Figure 6). Profiling revealed that IACS-9439 exhibits a high level of selectivity for CSF1R with no significant inhibition of other kinases; the outcome was also in agreement with the results of binding affinity determination that confirmed the high selectivity of IACS-9439 for CSF1R vs FLT3 (9,500-fold), cKIT (>17,000-fold), PDGFRα (1,900-fold) and PDGFRβ (450-fold). Notably, IACS-9439 (**1**) displayed superior selectivity when compared to BLZ945 in the above kinase panel. These results were also confirmed in a cellular context, where IACS-9439

not only showed improved potency in the pCSF1R cellular target engagement assay in comparison with BLZ945 (IC_{50} of 17 nM vs 155 nm), but also significantly higher selectivity for CSF1R against PDGFRβ (150-fold vs 3.7-fold) (Table 8).

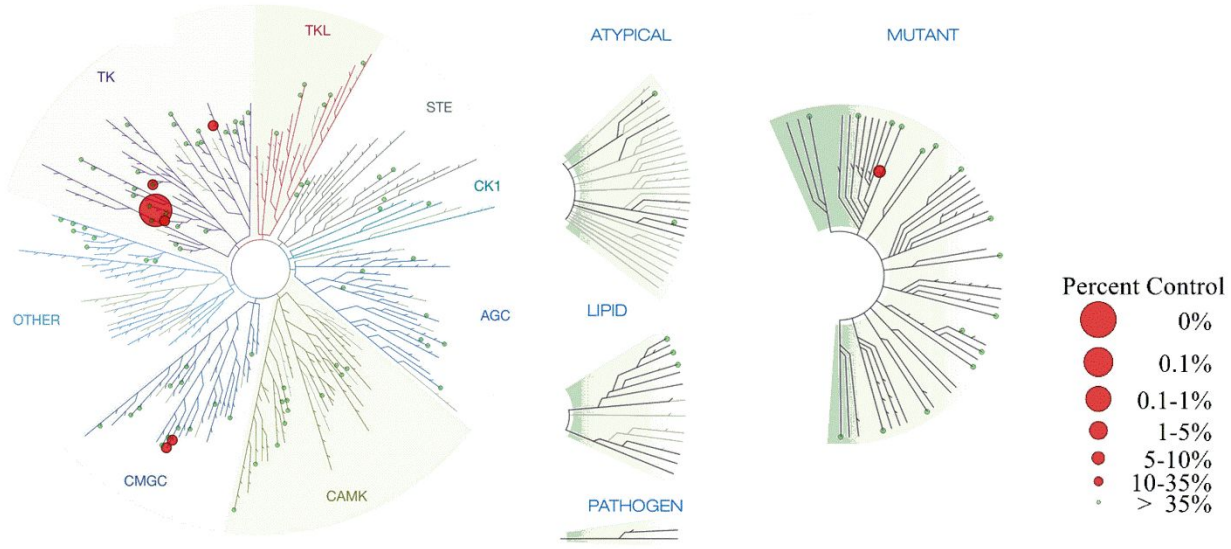


Figure 6. Selectivity profile of IACS-9439 (**1**) against a panel of 97 kinases (DiscoverX) at 1 uM test concentration. The larger the red circle, the greater the inhibition. One kinase with percent of control < 10: CSF1R. See also Table S3, Supporting information.

Table 8. Selectivity profile of IACS-9439 (**1**) and BLZ945 in binding affinity assays against type III kinases CSF1R, FLT3, cKIT, PDGFRα and PDGFRβ and in cellular pCSFR1 and pPDGFRβ target engagement assays.

Compound #	**BLZ945**	**IACS-9439 (1)**
Kd [nM] (Fold X) CSF1R	3.4	1
FLT3 [nM] (Fold X)	8700 (2552)	9,500 (9,500)
cKIT [nM] (Fold X)	3000 (882)	17,000 (17,000)
PDGFRα [nM] (Fold X)	210 (61)	1,900 (1,900)
PDGFRβ [nM] (Fold X)	46 (13)	450 (450)
Phospho-CSF1R [nM] (THP-1)	155	17

Phospho-PDGFRβ (HEK293) [nM] (Fold X)	579 (3.7)	3560 (150)

IACS-9439 (**1**) had low to moderate microsomal stability in rat and human microsomes and high microsomal turnover in mouse, dog and monkey microsomes. It had an acceptable free plasma concentration across species, with a free fraction (fu) of 6.5% in human (Table 9). IACS-9439 (**1**) possessed an suitable *in vivo* PK profile in rat and mouse, with moderate clearance, acceptable half-life and excellent oral bioavailabity. In dog and monkey, the compound had higher clearance and shorter half-life, as well as lower oral bioavailability.

Table 9. Microsomal and hepatic stablity, plasma protein binding and *in vivo* PK profile of IACS-9439.

Species	*In vitro* ADME properties of IACS-9439			*In vivo* pharmacokinetic data of IACS-9439				
	Plasma protein binding (%	Hepatocytes CLint,scaled [mL/min	Microsomes CLint,scaled [mL/min	Dose IV/PO [mg/kg]	CL [mL/min/kg] (IV)	Vss [L/kg]	$T_{1/2}$ [h] (IV)	Oral F [%]
Mouse	>99	21	91	0.3/10	14	1	2.2	100
Rat	97.9	15	38	0.3/3	13	3.4	1.7	81
Dog	95.2	64	293	0.3/3	30	3	1.37	37
Monkey	98.4	37	208	0.3/1	7.2	0.45	0.96	21
Human	93.5	32	14					

To confirm the biological activity of IACS-9439 (**1**) and compare it to compound **12** and BLZ945, we performed a series of *in vivo* experiments utilizing PANC02 and MC38 syngeneic tumor models. MC38 tumor bearing mice were treated with 20, 60 or 200 mg/kg BLZ945, compound **12** or IACS-9439 (**1**) for 10 days (n=3 mice/group) and similar dose dependent reductions in macrophages were observed by IHC and mRNAs using nanostring (Figure 7A) for each of the compounds.

To better understand the PK/PD relationship, mice bearing PANC02 tumors were treated with 10, 25, 50, 100 or 200 mg/kg IACS-9439 (**1**) or BLZ945for 5 days (n=4 mice/group) before harvesting tumor and plasma to measure changes in EMR1 (F4/80)

mRNA, as a measure of macrophages, and CD4 mRNA, as a measure of CD4 T-cells and compare these to the concentration of IACS-9439 (**1**) or BLZ945 in the plasma. For both compounds and both mRNAs, there was a clear PK/PD relationship with IACS-9439 (**1**) working at lower plasma concentrations (Figure 7B-C), a ~3 μM IC_{50} for IACS-9439 (**1**) compared to ~18 μM IC_{50} for BLZ945 for EMR1 mRNA and an ~18 μM IC_{50} for IACS-9439 (**1**) compared to an ~56 μM IC_{50} for BLZ945 for CD4 mRNA.

CSF1R inhibitors have been reported to promote macrophage polarization toward the M1 phenotype.[8] Consistent with these reports, we found, using flow cytometry, that treatment of mice bearing MC38 tumors with 10, 50 or 200 mg/kg IACS-9439 (**1**) or BLZ945for 10 days (n=10 mice/group) resulted in a dose dependent increase in the percentage of macrophages of the M1 class, with a concomitant dose dependent decrease in the percentage of macrophages in the tumors that were of the M2 phenotype (Figure 7D). This data is consistent with treatment of tumors with IACS-9439 (**1**) and the other CSF1R inhibitors shifting the tumor macrophage balance from a pro-tumor M2 phenotype to an anti-tumor M1 phenotype.

The functional consequence of this shift in macrophage biology, reduction of M2 macrophages, while increasing the percentage of M1 macrophages is highlighted by the observed identical decreases in the growth of MC38 tumors in mice treated with 200 mg/kg of IACS-9439 (**1**) or BLZ945 (Figure 7E) (n=10 mice/group). Taken together, the data indicate that IACS-9439 (**1**) is a potent selective inhibitor of CSF1R which impacts macrophage biology and the growth of tumors in immune competent environments.

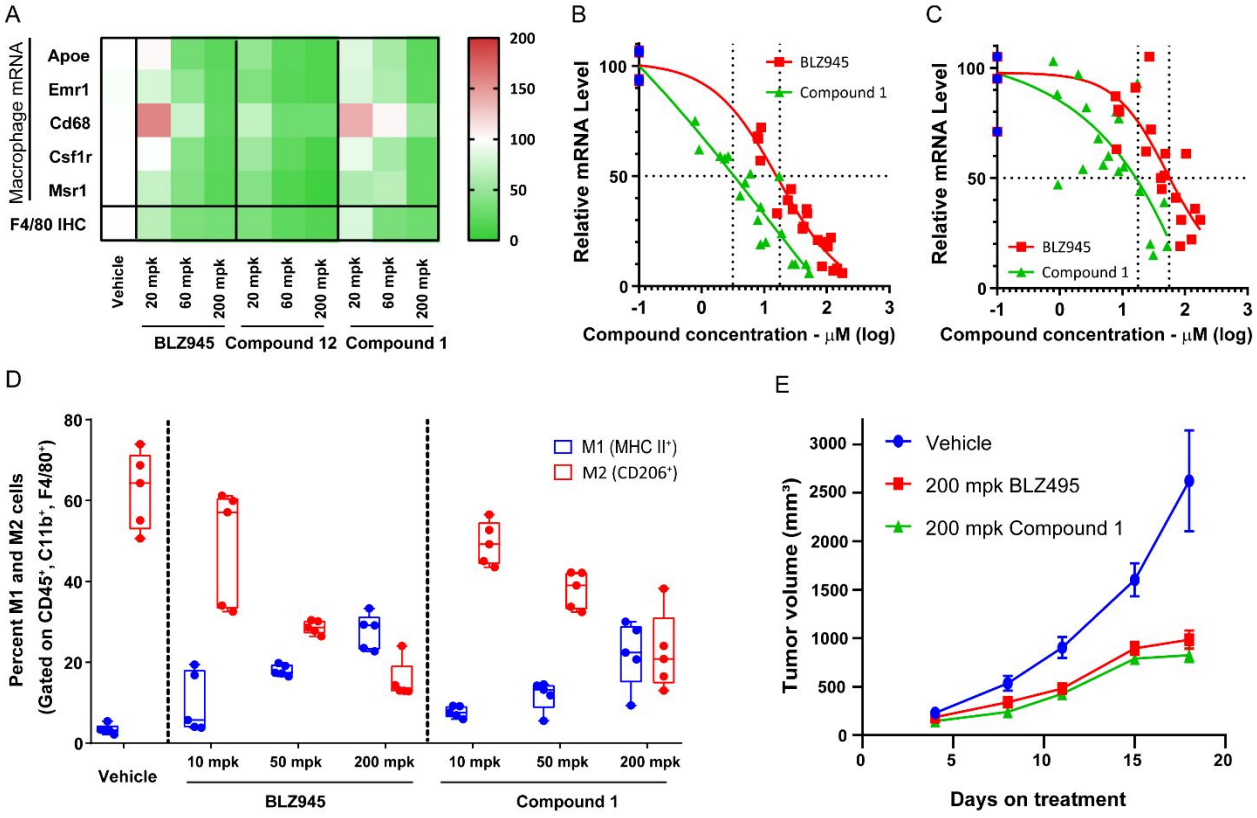


Figure 7. IACS-9439 (**1**) depletes tumor macrophages, promotes M1 macrophage and inhibits tumor growth. (A) Heat map of nanostring and IHC measuring relative

macrophage mRNAs or F4/80 protein levels from MC38 tumors treated daily for 10 days with vehicle or 20, 60 or 200 mg/kg of BLZ945, compound **12** or IACS-9439 (**1**) (n=3 mice/group). Note that 100 = no change (white) compared to vehicle treated mice. (B) Relative EMR1 (F4/80) or (C) CD4 mRNA from PANC02 tumors treated daily for 10 days with 10, 25, 50, 100 or 200 mg/kg BLZ945 or IACS-9439 (**1**) (n=4 mice/group). Tumor samples from CSF1Ri treated mice were normalized to vehicle treated mice and plotted in relation to the concentration of compound in the plasma. Note that vehicle was artificially set as 0.1 μM to enable curve fitting. (D) Flow cytometry depicting the percent of M1 (MHC II+) and M2 (CD206+) macrophages (CD45+, C11b+, F4/80+) in MC38 tumors from mice treated daily for 10 days with vehicle, 10, 50 or 200 mg/kg BLZ945 or IACS-9439 (**1**) (n=10 mice/group). (E) Growth of MC38 tumors implanted subcutaneously in mice and treated daily with vehicle, BLZ945 or 200 mg/kg IACS-9439 (**1**) (n=10 mice/group).

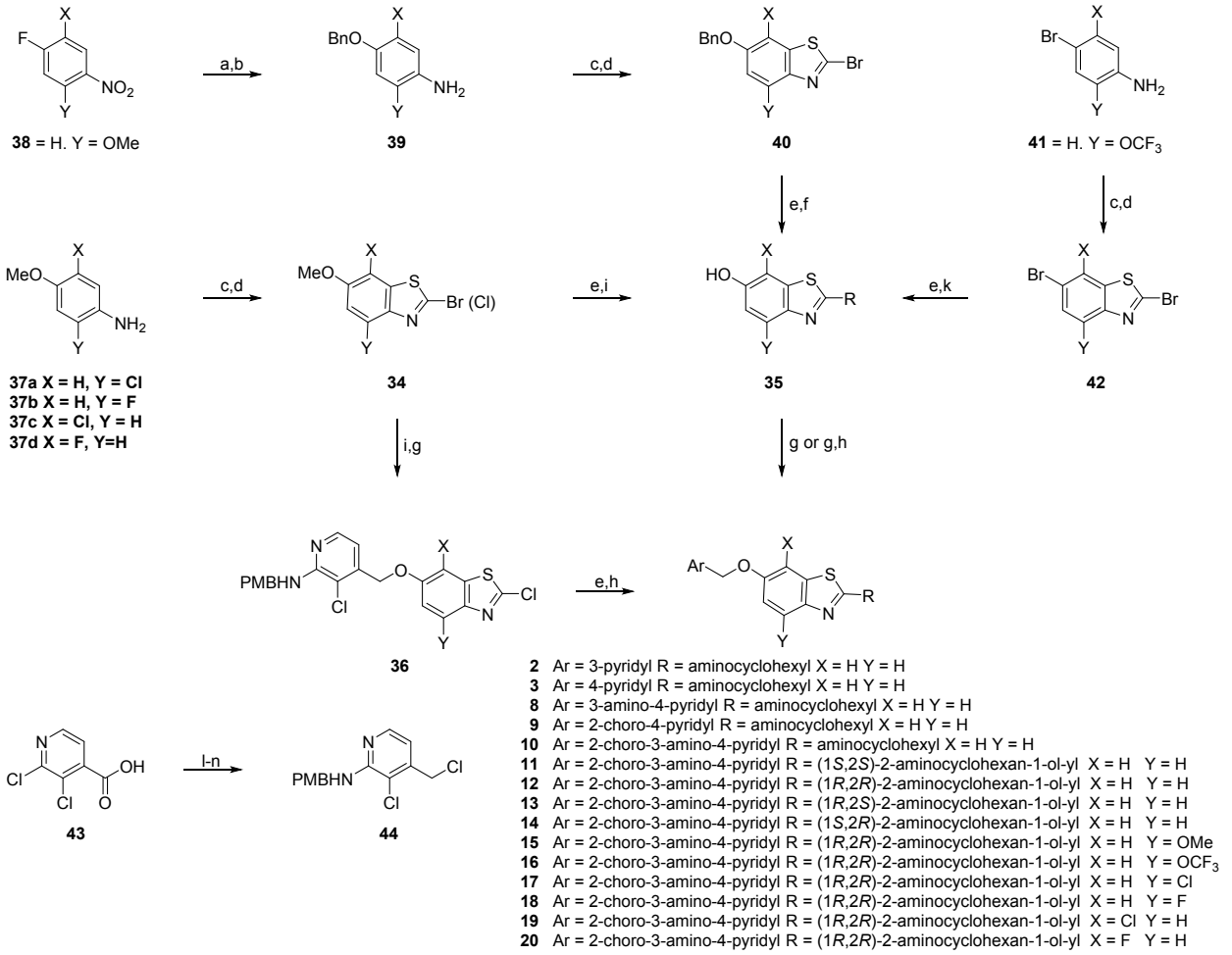
CHEMISTRY

Compounds **2**, **3** and **8-20** were synthesized as described in Scheme 1. Compound **2** was prepared starting from 2-bromo-6-methoxybenzo[d]thiazole **34** which was reacted with cyclohexylamine in the presence of K_2CO_3. Subsequent demethylation with BBr_3 afforded intermediate **35**, which was alkylated with 3-(chloromethyl)pyridine to afford compound **2**. Compound **3** was prepared in a similar manner, utilizing 4-(chloromethyl)pyridine as the alkylating agent in the last step. Compound **8** was obtained following a similar route, by alkylating intermediate **35** with tert-butyl 4-(chloromethyl)pyridin-2-ylcarbamate, followed by removal of the Boc group by TFA to provide final compound **8**. Alkylation of intermediate **35** with 3-chloro-4-(chloromethyl)pyridine afforded compound **9**. To obtain compound **10**, intermediate **35** was reacted with 3-chloro-4-(chloromethyl)-N-(4-methoxybenzyl)pyridin-2-amine **44**, which was prepared in three steps from 2,3-dichloroisonicotinic acid **43**. Final deprotection with TFA provided the desired compound **10**. Compounds **11** and **12** were synthesized in a similar fashion. 2-Bromo-6-methoxybenzo[d]thiazole **34** was reacted with (1S,2S)-2-aminocyclohexan-1-ol or (1R,2R)-2-aminocyclohexan-1-ol, respectively, and the resulting product was demethylated using BBr_3 to provide intermediate **35**. Subsequent alkylation with 3-chloro-4-(chloromethyl)-N-(4-methoxybenzyl)pyridin-2-amine **44** and deprotection with TFA provided the final products **11** and **12**. To synthesize

compounds **13** and **14**, 2-chloro-6-methoxybenzo[d]thiazole **34** was demethylated and alkylated with 3-chloro-4-(chloromethyl)-N-(4-methoxybenzyl)pyridin-2-amine **44.** S_NAr reaction with (1R,2S)-2-aminocyclohexan-1-ol or (1S,2R)-2-aminocyclohexan-1-ol and subsequent deprotection provided the desired products **13** and **14**. Compound **15** was synthesized starting from 4-fluoro-2-methoxy-1-nitrobenzene **38**, which was reacted with BnOH under S_NAr conditions and subsequently reduced to the corresponding aniline derivative **39**. The benzothiazole ring was formed by treating compound **39** with KSCN and $CuSO_4$ in MeOH. Subsequent reaction with *t*-BuONO and $CuBr_2$ led to intermediate **40**. S_NAr reaction of **40** with (1R,2R)-2-aminocyclohexan-1-ol followed by deprotection with TFA provided the intermediate **35**. Alkylation of **35** with 3-chloro-4-(chloromethyl)-N-(4-methoxybenzyl)pyridin-2-amine **44** and subsequent deprotection led to analog **15**. Compound **16** was synthesized in a slightly different manner. Thiazole derivative **42** was prepared by reacting 4-bromo-2-(trifluoromethoxy)aniline **41** with KSCN in the presence of Br_2 and AcOH and subsequent treatment of the product with *t*-BuONO and $CuBr_2$. S_NAr reaction of **42** with (1R,2R)-2-aminocyclohexan-1-ol followed by introduction of the 4,4,5,5-tetramethyl-1,3,2-dioxaborolan-2-yl functionality by $Pd(dppf)Cl_2$ catalyzed

coupling with B_2Pin_2 and subsequent treatment with H_2O_2 led to phenol intermediate **35**. Alkylation and subsequent deprotection provided the desired product **16**. Compounds **17-20** were prepared according to the sequence as it was described for compound **12**, utilizing the appropriately substituted 2-bromo-6-methoxybenzo[d]thiazole intermediates **34**, which in turn could be obtained from substituted anilines **37a-d** in two steps.

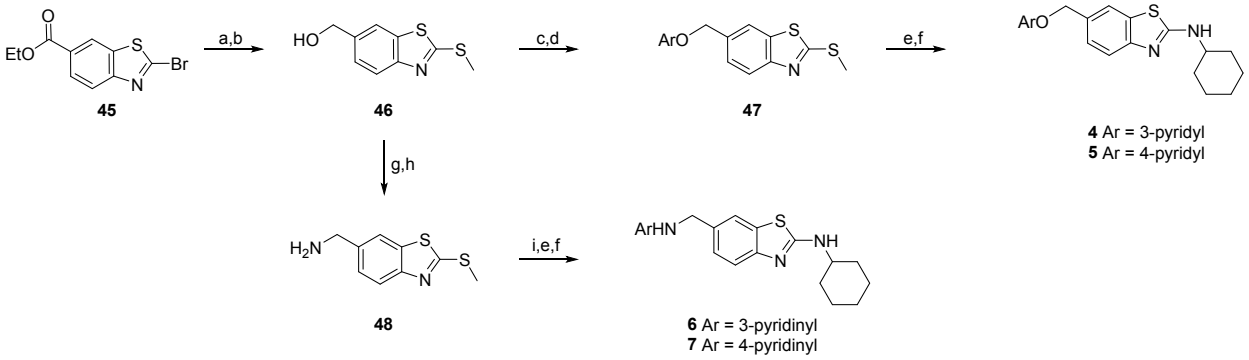
Scheme 1. Chemical synthesis of compounds **2, 3 and 8-20.**



Reagents and conditions: a) BnOH, NaH, 0°C, DMF, 99%; b) N_2H_4-H_2O, Raney-Ni, rt, MeOH, 88%; c) KSCN, $CuSO_4$, 90°C, MeOH, 37%; or KSCN, Br_2, HOAc, 0°C to rt, 4h, AcOH, 12.5-78%; d) t-BuONO, $CuBr_2$, 0°C to rt, MeCN, 30-53%; e) amine, K_2CO_3, DMSO, 100°C, 81%; or amine, DIPEA, DMF, 110°C, 78-100%; or amine, 110°C, 79%; or amine, Na_2CO_3, NMP, 125°C, 66-70%; f) TFA, 110°C, 100%; g) $ArCH_2Cl$, K_2CO_3, DMF, 80°C, 42-88%; or $ArCH_2Cl$, Cs_2CO_3, DMF, 80°C, 27-80%; h) TFA, rt, quant.; i) BBr_3, 0°C to rt, DCM, 78-90%; j) B_2Pin_2, KOAc, $Pd(dppf)Cl_2$, 100°C, 1,4-dioxane, quant.; k) H_2O_2, rt, 1,4-dioxane, 60%; l) BH_3-THF; 60°C, 45%; m) 4-methoxybenzyl amine, 150°C, 64%; n) $SOCl_2$, rt, 2h, DCM, 94%.

Compounds **4-7** were synthesized as described in Scheme 2. 2-Bromobenzo[d]thiazole-6-carboxylate **45** was reduced with DIBAL-H and subsequently reacted with sodium thiomethoxide to provide intermediate **46**. Mesylate formation and reaction with pyridin-3-ylmethanol or pyridin-4-ylmethanol, respectively, led to intermediate **47**, which, upon oxidation with mCPBA, and subsequent S_NAr reaction provided compounds **4** and **5**. To obtain compounds **6** and **7**, intermediate **46** was treated with DPPA and the resulting azide was reduced to the corresponding amine **48** using Lindlar's catalyst. Amine **48** was reacted with 3-iodopyridine or 4-iodopyridine, respectively. Oxidation of intermediate **48** followed by S_NAr reaction led to compounds **6** and **7**.

Scheme 2. Chemical synthesis of compounds **4-7.**

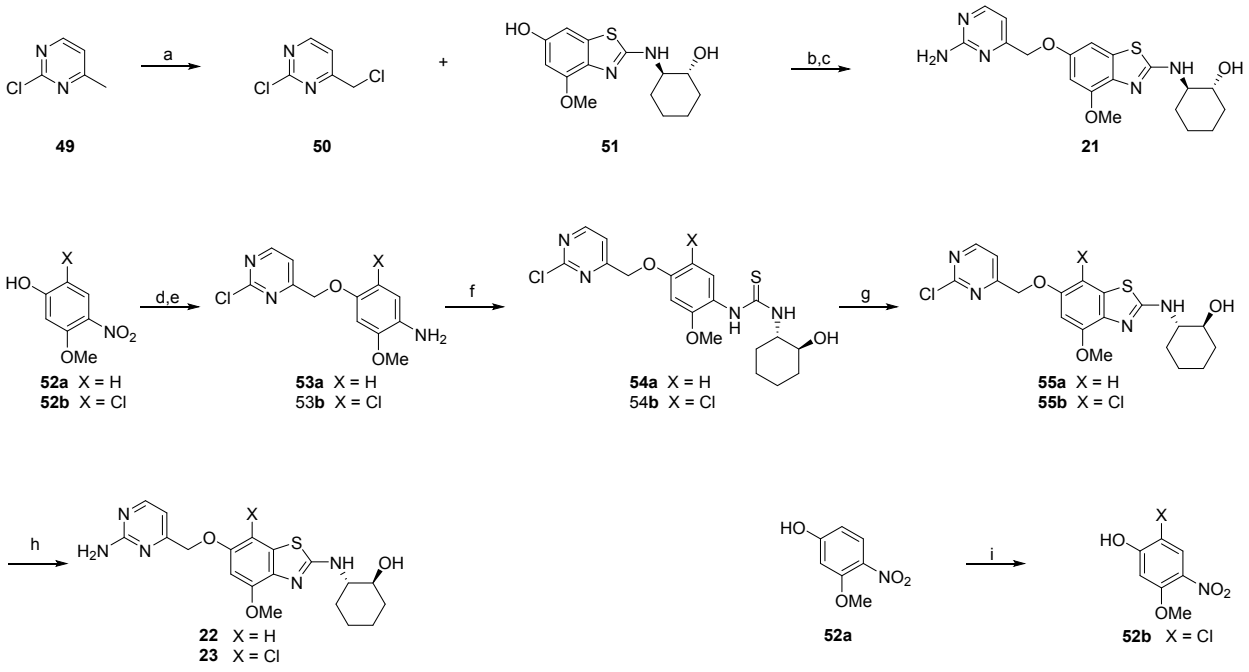


Reagents and conditions: a) DIBAL-H, THF, 0°C-rt 4h, 98%; b) MeSNa, DMF,12h, 71%; c) MsCl, DIPEA, DMF. DCM, 0°C-rt, 12h, quant.; d) pyridin-3-ylmethanol or pyridin-4-ylmethanol, tBuOK, DMA, 100°C, 2h, quant.; e) *m*-CPBA, DCM, 2h, quant.; f) cyclohexanamine, DIPEA, DMA,120°C 2h, 18%; g) DPPA, DBU, THF, rt, 3h, 92%; h)

Lindlar Cat., H_2, EtOH, 2h; i) 3-iodopyridine or 4-iodopyridine, Cu, CsOAc, DMSO, 90°C, 2h, 78%.

Compounds **21-23** were synthesized as described in Scheme 3. 2-Chloro-4-methylpyrimidine **49** was treated with NCS in the presence of AIBN in CCl_4. The resulting chloromethyl pyridine derivative **50** was reacted with benzothiazole derivative **51**. Subsequent S_NAr reaction with NH_3-H_2O provided the desired product **21**. To synthesize compound **22** in an efficient manner, a novel route was developed. 3-Methoxy-4-nitrophenol **52a** was reacted with 2-chloro-4-(chloromethyl)pyrimidine **50** and the nitro group of the resulting product was reduced using Pt/C to afford intermediate **53a**. Reacting compound **53a** with di(1H-imidazol-1-yl)methanethione and subsequently with (1S,2S)-2-aminocyclohexanol led to intermediate **54a**, which upon treatment with $BnMe_3NBr_3$ and BSTFA was cyclized to the desired benzothiazole. Final S_NAr reaction with NH_3 yielded compound **22**. Compound **23** was prepared in a similar manner, starting out from 2-chloro-5-methoxy-4-nitrophenol **52b** that was synthesized starting from 3-methoxy-4-nitrophenol **52a** by treatment with NCS.

Scheme 3. Chemical synthesis of compounds **21-23.**

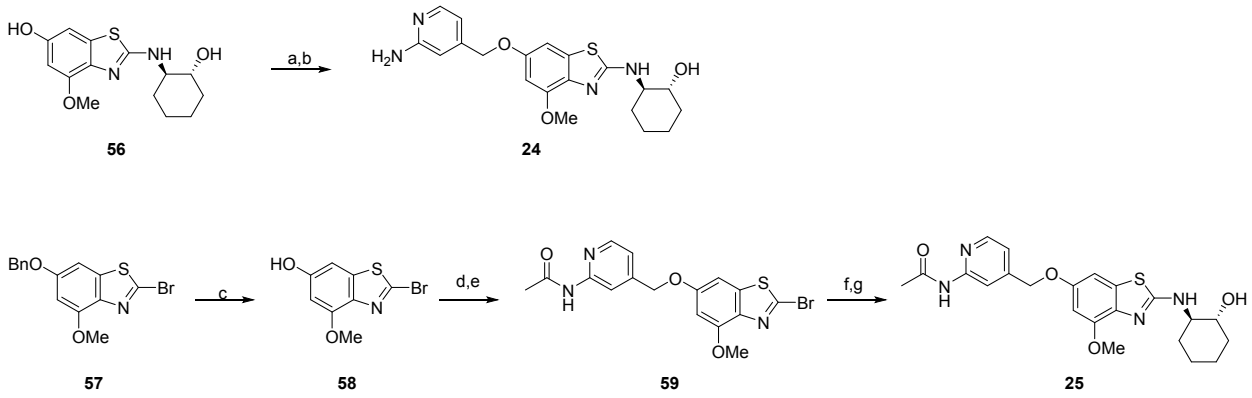


Reagents and conditions: a) NH_3, H_2O 90°C, 3h, 49%; b) AIBN, NCS, CCl_4, 18h, 67%; c) **50**, Cs_2CO_3, rt, 4h, DMF, 8%; d) **50**, K_2CO_3, rt, 20h, DMF, 93%; e) 10% Pt-C, rt, 2h, THF-MeOH, (5:1), 97%; f) di(1H-imidazol-1-yl)methanethione, (1S,2S)-2-aminocyclohexan-1-ol, rt, 4h, DCM, 96%; g) $BnMe_3NBr_3$, BSTFA, rt, 20 min, DCM, 95%; h) NH_3, (7M, in MeOH), 120°C, 4h, 64%; i) NCS, 50°C, 2h, DCM, quant.

Compounds **24** and **25** were synthesized as described in Scheme 4. Intermediate **56** was alkylated with tert-butyl (4-(chloromethyl)pyridin-2-yl)carbamate and was subsequently deprotected to provide the desired product **24**. Compound **25** was prepared

following a modified sequence, starting with intermediate **57** that was deprotected to afford benzothiazole derivative **58**. Mitsunobu reaction with tert-butyl (4-(hydroxymethyl)pyridin-2-yl)carbamate yielded intermediate **59**. Removal of the Boc group by TFA, followed by acylation with Ac_2O in the presence of pyridine, and subsequent S_NAr reaction with (1R,2R)-2-aminocyclohexan-1-ol provided the desired product **25**.

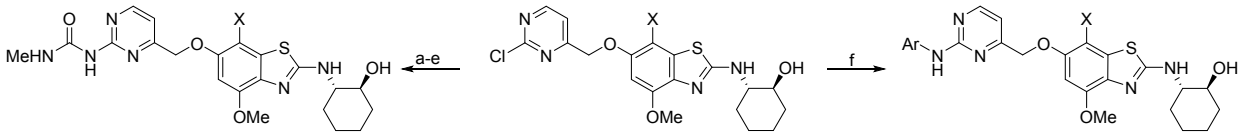
Scheme 4. Chemical synthesis of compounds **24** and **25.**



Reagents and conditions: a) tert-butyl (4-(chloromethyl)pyridin-2-yl)carbamate, Cs_2CO_3, 80°C, 2h, DMF, 17%; b) TFA, rt, 16h, 35%; c) TFA, 65°C, 24h 40h, 24%; d) tert-butyl (4-(hydroxymethyl)pyridin-2-yl)carbamate, Ph_3P, DtBAD, rt, 16h, THF, 93%; e) TFA, rt, 3h, DCM, 60%; f) Ac_2O, pyr, 60°C, 1h, DMF, 88%; g) (1R,2R)-2-aminocyclohexan-1-ol DIPEA, DMA, 100°C, 12h, 38%.

Compounds **26-33** were prepared as shown in Scheme 5. Compound **26** was prepared from intermediate **55a** in five steps. First, the secondary alcohol was protected with a TBS group. Subsequent S_NAr reaction with ammonia, followed by treatment phenylchloroformate and methylamine, and final deprotection yielded compound **26**. Compounds **1** and **26-31** were prepared from intermedate **55a** in an S_NAr reaction with the appropriate aromatic amine. Compound **32** was prepared from intermediate **55b**, in a similar manner, via an S_NAr reaction with 1-methyl-1H-pyrazol-4-amine to provide the desired product.

Scheme 5. Chemical synthesis of compounds **26-33**.



26 X = H

55a X = H
55b X = Cl

1 X = H, Ar = 1-methyl-1H-pyrazol-4-amine
27 X = H, Ar = 1-methyl-1H-pyrazol-3-amine
28 X = H, Ar = 1H-pyrazol-4-amine
29 X = H, Ar = 1H-pyrazol-3-amine
30 X = H, Ar = 4-amino isoxazole
31 X = H, Ar = 1-methyl-1H-1,2,3-triazol-4-amine
32 X = H, Ar = 3-pyridyl
33 X = Cl Ar = 1-methyl-1H-pyrazol-4-amine

Reagents and conditions:a) TBSCl, imid., DMF, 12h, rt, 91%; b) NH_3 in *i*PrOH (2M),120°C, MW, 8h, 72%; c) phenylchloroformate, Py, DMAP (Cat.), 3h, rt; d) $MeNH_2$, 12h, rt, 51% for two steps; e) TBAF, 12h, rt, 40%; f) i. 1-methyl-1H-pyrazol-4-amine DIPEA, 120°C, 10h, DMA, 46% (**1**); ii. 1-methyl-1H-pyrazol-3-amine, DIPEA, 120°C, 5h, 2-Propanol, MW, 15%; (**27**); iii.1H-pyrazol-4-amine, TEA, 120°C, 2h, DMSO, MW, 44% (**28**); iv. 1H-pyrazol-3-amine, $Pd_2(dba)_3$, Xantphos, Cs_2CO_3, dioxane, 110°C, 2h, 14% (**29**); v. 4-amino isoxazole hydrochloride, TEA, 120°C, 2h, DMSO, MW, 63% (**30**); vi.1-methyl-1H-1,2,3-triazol-4-amine, TsOH, dioxan, 110°C, 12h, 10% (**31**); vii. 1-methyl-1H-pyrazol-3-amine, DIPEA, 110°C, 18h, , 5% (**32**).

CONCLUSIONS

Rational design and iterative, hypothesis driven potency and properties optimization allowed us to develop a highly potent, exquisitely selective and orally bioavailable CSF1R inhibitor, IACS-9439 (**1**). Deep analysis of the active site in the DGF-out conformation of kinases CSF1R, cKit, FLT3, PDGFRα and PDGFRβ enabled us to rapidly design analogs of the initial lead, compound **12**, with drastically improved selectivity. Comprehensive evaluation of the selectivity of these inhibitors was enabled by utilizing a suite of assays including kinase binding as well as enzymatic assays for the above kinases. To evaluate selectivity in a cellular context, CSF1R and PDGFRβ cellular target engagement assays were implemented. Subsequent hypothesis-driven properties optimization of the highly

selective CSF1R inhibitor **15** led to analogs with improved DMPK properies. Finally, a significant boost in potency was achieved by introducing a substituent onto the hinge binding motif, resulting in the identification of IACS-9439 (**1**). The excellent potency, exquisite CSF1R selectivity and *in vivo* PK profile of IACS-9439 (**1**) allowed us to utilize it as an *in vivo* tool to probe CSF1R-mediated biology. We demonstrated that treatment with IACS-9439 (**1**) led to dose-dependent reduction of macrophages and promoted macrophage polarization toward the M1 phenotype in the syngeneic tumor model, and led to tumor growth inhibition in MC38 and PANC01 tumor models. We also developed a novel, highly efficient and scalable synthetic route for the preparation of 4-methoxy substituted benzothiazole analogs and IACS-9439 (**1**).

EXPERIMENTAL SECTION

Synthetic methods: The inhibitors described were synthesized by employing standard chemical transformations. Starting materials and reagents were purchased from commercial suppliers such as Sigma-Aldrich, Alfa Aesar, TCI, or Acros and will be used without further purification unless otherwise indicated. Anhydrous solvents (e.g., THF,

DMF, DMA, DMSO, MeOH, DCM, toluene) were purchased from Sigma-Aldrich and used directly. Purification of inhibitors were performed by column chromatography utilizing a Biotage system applying Biotage SNAP columns with Biotage KP-Sil silica or Biotage Zip Si columns with Biotage KP-Sil silica or a Teledyne ISCO system with RediSep Rf normal phase silica cartridges. Other inhibitors were purified by preparative HPLC using a Waters Autopurify system with a Waters Xbridge Prep C18 5 µm OBD, 19 mm × 150 mm or 50 mm × 100 mm column and SQ detector mass spectrometer with ESI ionization. The identity of all compounds with reported biological activity was confirmed by NMR spectroscopy and Low Resolution Mass Spectrometry, and for selected analogs, High Resolution Mass Spectrometry. Purity of all compounds with reported biological activity was $\geq$ 95% and was determined by Ultra Performance Liquid Chromatography (UPLC). NMR spectra were recorded on Bruker instruments operating at 300, 500, or 600 MHz. NMR spectra were obtained as $CDCl_3$, CD_3OD, D_2O, $(CD_3)_2SO$, $(CD_3)_2CO$, C_6D_6, or CD_3CN solutions (reported in ppm), using tetramethylsilane (0.00 ppm) or residual solvent ($CDCl_3$, 7.26 ppm; CD_3OD, 3.31 ppm; D_2O, 4.79 ppm; $CD_3)_2SO$, 2.50 ppm; $(CD_3)_2CO$, 2.05 ppm; C_6D_6, 7.16 ppm; CD_3CN, 1.94 ppm) as the reference standard. Low-resolution

mass spectral were obtained on either a Waters H class UPLC with a Waters Acquity UPLC BEH C18 1.7 μm, 2.1 mm × 50 mm column, UV detection between 200 and 400 nm, evaporating light scattering detection, and a SQ detector mass spectrometer with ESI ionization or a Water I class UPLC with a Waters Acquity UPLC CSH C18 1.7 μm, 2.1 mm × 50 mm column, UV detection at 254 and 290 nm, evaporating light scattering detection, and a SQ detector 2 mass spectrometer with ESI ionization. High-resolution mass spectra were obtained on a Waters Acquity I-Class UPLC coupled to a LTQ-Orbitrap Elite mass spectrometer. The injection volume was 5 μL. Chromatographic separation was performed on a Waters Acquity UPLC BEH C18 1.7 μm, 2.1 mm × 50 mm column, at a flow rate of 0.5 mL/min. The mobile phases were 0.1% Acetic Acid in Water (solvent A) and 0.1% Acetic acid in acetonitrile (solvent B). The gradient had a total run time of 18 minutes and was as follows: 0-2 minutes 5% B; 2-12 minutes from 5% to 65%B; 12-14 minutes from 65% to 95% B; 14-16 minutes at 95%B; 16-16.1 minutes from 95% to 5%B and 16.1-18 minutes at 5% B. The column temperature was kept at 40 °C. The samples were analyzed using the positive electrospray ionization (ESI) mode. The ESI source temperature was set at 375 °C, the capillary temperature at 320 °C and the

electrospray voltage at 4.1 kV. Sheath and auxiliary gas were 45 arbitrary unit and 10 arbitrary unit, respectively.

Synthesis of N-cyclohexyl-6-(pyridin-3-ylmethoxy)benzo[d]thiazol-2-amine (2)

Step 1: N-cyclohexyl-6-methoxybenzo[d]thiazol-2-amine. A mixture of 2-bromo-6-methoxybenzo[d]thiazole (2.0 g, 8.2 mmol), cyclohexanamine (1.1 g, 11 mmol) and K_2CO_3 (2.3 g, 16 mmol) in DMSO (15 mL) was heated at 100°C for 22 h. The reaction mixture was cooled to rt, diluted with water (25 mL) and extracted with EtOAc (3 x 30 mL). The combined organic layers were dried over $MgSO_4$ and concentrated under reduced pressure. The residue was purified by flash column chromatography (10% to 30% EtOAc in Hexanes) to provide N-cyclohexyl-6-methoxybenzo[d]thiazol-2-amine (1.7 g, 81%) as a yellow solid. MS (ES+) $C_{14}H_{18}N_2OS$ requires: 262, found: 263$[M+H]^+$.

Step 2: 2-(cyclohexylamino)benzo[d]thiazol-6-ol. To a solution of N-cyclohexyl-6-methoxybenzo[d]thiazol-2-amine (1.7 g, 6.7 mmol) in DCM (30 mL), at 0°C, BBr_3 (4.2 g, 17 mmol) was added slowly and the resulting mixture was stirred at rt for 3 h. The reaction mixture was slowly quenched with ice-water. $NaHCO_3$ (4.2 g, 51 mmol) was

added. The precipitate was filtered to afford 2-(cyclohexylamino)benzo[d]thiazol-6-ol (1.3 g, 78%) as a beige solid. MS (ES+) $C_{13}H_{16}N_2OS$ requires: 248, found: 249$[M+H]^+$.

Step 3: N-cyclohexyl-6-(pyridin-3-ylmethoxy)benzo[d]thiazol-2-amine). A mixture of 2-(cyclohexylamino)benzo[d]thiazol-6-ol (75 mg, 0.30 mmol), 3-(chloromethyl)pyridine hydrochloride (59 mg, 0.36 mmol) and K_2CO_3 (83 mg, 0.60 mmol) in DMF (1 mL) was heated to 80°C and was stirred at this temperature for 16 h. The mixture was purified by preparative HPLC (Mobile phase: A = 0.1% TFA/H_2O, B = 0.1% TFA/MeCN to provide N-cyclohexyl-6-(pyridin-3-ylmethoxy)benzo[d]thiazol-2-amine (89 mg, 88%) as a brown solid. MS (ES+) $C_{19}H_{21}N_3OS$ requires: 339, found: 340[M+H]+; ^{1}H NMR (500 MHz, d_6-DMSO) δ 8.84 (s, 1H), 8.72 (d, J = 4.3 Hz, 1H), 8.63 (s, 1H), 8.22 (d, J = 7.7 Hz, 1H), 7.73 (dd, J = 7.3, 5.7 Hz, 1H), 7.50 (d, J = 1.9 Hz, 1H), 7.35 (d, J = 8.7 Hz, 1H), 7.01 (dd, J = 8.8, 2.0 Hz, 1H), 5.23 (s, 2H), 3.73 – 3.60 (m, 1H), 2.02 – 1.93 (m, 2H), 1.77 – 1.68 (m, 2H), 1.62 – 1.54 (m, 1H), 1.39 – 1.17 (m, 5H).

Synthesis of N-cyclohexyl-6-(pyridin-4-ylmethoxy)benzo[d]thiazol-2-amine (3)

Step 1: N-cyclohexyl-6-(pyridin-4-ylmethoxy)benzo[d]thiazol-2-amine. A mixture of 2-(cyclohexylamino)benzo[d]thiazol-6-ol (75 mg, 0.30 mmol), 4-(chloromethyl)pyridine

hydrochloride (59 mg, 0.36 mmol) and K_2CO_3 (83 mg, 0.60 mmol) in DMF was heated at 80°C for 2 h. The residue was purified by preparative HPLC (Mobile phase: A = 0.1% TFA/H_2O, B = 0.1% TFA/MeCN to give N-cyclohexyl-6-(pyridin-4-ylmethoxy)benzo[d]thiazol-2-amine (79 mg, 78%) as a beige solid. MS (ES+) $C_{19}H_{21}N_3OS$ requires: 339, found: 340[M+H]+; ^{1}H NMR (500 MHz, *d*$_6$-DMSO) δ 8.81 (s, 1H), 7.81 (d, *J* = 2.5 Hz, 2H), 7.44 (d, *J* = 2.6 Hz, 1H), 7.33 (d, *J* = 8.8 Hz, 1H), 6.97 (dd, *J* = 8.8, 2.6 Hz, 1H), 5.34 (s, 2H), 3.70 – 3.61 (m, 1H), 2.02 – 1.93 (m, 2H), 1.77 – 1.68 (m, 2H), 1.62 – 1.55 (m, 1H), 1.38 – 1.18 (m, 5H).

Synthesis of 6-((2-aminopyridin-4-yl)methoxy)-N-cyclohexylbenzo[d]thiazol-2-amine (8):

Step 1 : tert-butyl 4-(chloromethyl)pyridin-2-ylcarbamate. To a solution of tert-butyl 4-(hydroxymethyl)pyridin-2-ylcarbamate (67 mg, 0.30 mmol) in DCM (2 mL) was added $SOCl_2$ (10 drops). The reaction mixture was stirred at rt for 2 h. The mixture was concentrated to provide tert-butyl 4-(chloromethyl)pyridin-2-ylcarbamate as a yellow solid which was used directly in the next step without further purification. MS (ES+) $C_{11}H_{15}ClN_2O_2$ requires: 242, found: 243[M+H]+.

Step 2: 6-((2-aminopyridin-4-yl)methoxy)-N-cyclohexylbenzo[d]thiazol-2-amine. A mixture of 2-(cyclohexylamino)benzo[d]thiazol-6-ol (50 mg, 0.20 mmol) and Cs_2CO_3 (163 mg, 0.50 mmol) in DMF (2 mL) was stirred at rt for 2 h. *Tert*-butyl 4-(chloromethyl)pyridin-2-ylcarbamate (73 mg, 0.30 mmol) was added. The reaction mixture was stirred at 120°C for 4 h. The residue was purified by preparative-HPLC (Mobile phase: A = 0.1% NH_4HCO_3/H_2O, B = MeCN; Gradient: B = 5 - 95%; 12 min; Column: C18 to provide 6-((2-aminopyridin-4-yl)methoxy)-N-cyclohexylbenzo[d]thiazol-2-amine (29 mg, 28%) as a beige solid. MS (ES+) $C_{19}H_{22}N_4OS$ requires: 354, found: 355 [M+H]+; ^{1}H NMR (400 MHz, d_4-Methanol) δ 7.88 (dd, *J* = 5.4, 0.7 Hz, 1H), 7.33 (d, *J* = 8.8 Hz, 1H), 7.24 (d, *J* = 2.5 Hz, 1H), 6.94 (dd, *J* = 8.8, 2.6 Hz, 1H), 6.72 – 6.65 (m, 2H), 5.02 (s, 2H), 3.69-3.66 (m, 1H), 2.10-2.08 (m, 2H), 1.86 – 1.77 (m, 2H), 1.70-1.68 (m, 1H), 1.47-1.42 (m, 2H), 1.37 – 1.25 (m, 3H).

Synthesis of 6-((3-chloropyridin-4-yl)methoxy)-N-cyclohexylbenzo[d]thiazol-2-amine (9)

Step 1: Synthesis of (3-chloropyridin-4-yl)methanol. To a solution of 3-chloroisonicotinaldehyde (660 mg, 4.66 mmol) in MeOH (20 mL) was added $NaBH_4$ (530 mg, 143 mmol) 0°C. The reaction mixture was stirred at 0°C for 2 h. The reaction was quenched with NH_4Cl (sat. aq.), diluted with water (80 mL) and extracted with DCM/MeOH 20:1 (3 x 30 mL). The organic layer was dried over $MgSO_4$, and concentrated under reduced pressure to provide (3-chloropyridin-4-yl)methanol as a light yellow solid (520 mg, 78%), which was used in the next step without further purification. MS (ES+) C_6H_6ClNO requires: 143, found: 144[M+H]+.

Step 2: Synthesis of 3-chloro-4-(chloromethyl)pyridine. To a solution of (3-chloropyridin-4-yl)methanol (520 mg, 3.63 mmol) in DCM (4 mL) was added $SOCl_2$ (2.0 mL, 27 mmol). The reaction mixture was stirred at rt for 2 h and concentrated under reduced pressure to provide 3-chloro-4-(chloromethyl)pyridine as a tan solid, which was used in the next step without further purification. MS (ES+) $C_6H_5Cl_2N$ requires: 161, found: 162[M+H]+.

Step 3: Synthesis of 6-((3-chloropyridin-4-yl)methoxy)-N-cyclohexylbenzo[d]thiazol-2-amine. A mixture of 2-(cyclohexylamino)benzo[d]thiazol-6-ol (40 mg, 0.16 mmol) and K_2CO_3 (110 mg, 0.797 mmol) in DMF (2 mL) was stirred at rt for 1 h. 3-Chloro-4-

(chloromethyl)pyridine (78 mg, 0.48 mmol) was added. The reaction mixture was stirred at rt for 48 h, purified by prep-HPLC (Mobile phase: A = 0.1% NH_4HCO_3/H_2O, B = MeCN; Gradient: B = 5 - 95%; 12 min; Column: C18) to provide 6-((3-chloropyridin-4-yl)methoxy)-N-cyclohexylbenzo[d]thiazol-2-amine (25 mg, 42%) as a yellow solid. MS (ES+) $C_{19}H_{20}ClN_3OS$ requires: 373, found: 374 [M+H]+; ^{1}H NMR (500 MHz, *d*$_4$-MeOD) δ 8.59 (s, 1H), 8.51 (d, *J* = 5.0 Hz, 1H), 7.70 (d, *J* = 5.0 Hz, 1H), 7.34 (dd, *J* = 23.0, 5.7 Hz, 2H), 7.00 (dd, *J* = 8.8, 2.6 Hz, 1H), 5.25 (s, 2H), 3.84 – 3.52 (m, 1H), 2.13 – 2.04 (m, 2H), 1.87 – 1.75 (m, 2H), 1.69 (dd, *J* = 9.3, 3.8 Hz, 1H), 1.53 – 1.22 (m, 5H).

Synthesis of 6-((2-amino-3-chloropyridin-4-yl)methoxy)-N-cyclohexylbenzo[d]thiazol-2-amine (10)

Step 1: (2,3-dichloropyridin-4-yl)methanol. A mixture of 2,3-dichloroisonicotinic acid (960 mg, 5.00 mmol) and BH_3•THF (1.0 M, 25 mL, 25 eq) was heated at 60°C for 4 h. The reaction mixture was cooled to rt, MeOH (5 mL) was added, and the volatiles were removed under reduced pressure. The mixture was diluted with water (50 mL) and extracted with DCM (3 x 50 mL). The combined organic phases were washed with brine

(1 x 100 mL), dried over Na_2SO_4, filtered and concentrated to provide (2,3-dichloropyridin-4-yl)methanol (400.5 mg, 45%) as a white solid. MS (ES+) $C_6H_5Cl_2NO$ requires: 178, found: 179 $[M+H]^+$.

Step 2: (3-chloro-2-(4-methoxybenzylamino)pyridin-4-yl)methanol. A mixture of (2,3-dichloropyridin-4-yl)methanol (200 mg, 1.12 mmol) and (4-methoxyphenyl)methanamine (1.0 mL, 7.6 mmol) was heated at 150°C for 4 h. The residue was purified by mass-triggered preparative HPLC (Mobile phase: A = 0.1% TFA/H_2O, B = 0.1% TFA/MeCN; Gradient: B = 10 - 90%; 12 min; Column: C18) to provide (3-chloro-2-(4-methoxybenzylamino)pyridin-4-yl)methanol (201 mg, 64%) as a white solid. MS (ES+) $C_{14}H_{15}ClN_2O_2$ requires: 278, found: 279 $[M+H]^+$. ^{1}H NMR (500 MHz, DMSO-d_6) δ 7.94 (d, J = 5.3 Hz, 1H), 7.24 (d, J = 8.7 Hz, 2H), 6.91 – 6.72 (m, 3H), 4.52 (d, J = 12.7 Hz, 4H), 3.71 (s, 3H).

Step 3: Synthesis of 3-chloro-4-(chloromethyl)-N-(4-methoxybenzyl)pyridin-2-amine. A mixture of (3-chloro-2-(4-methoxybenzylamino)pyridin-4-yl)methanol (50 mg, 0.18 mmol) and $SOCl_2$ (2.0 mL, 27 mmol) in DCM (2 mL) was stirred at rt for 2 h. The volatiles were removed under reduced pressure to provide 3-chloro-4-(chloromethyl)-N-(4-

methoxybenzyl)pyridin-2-amine (51 mg, 94%) as a yellow solid. MS (ES+) $C_{14}H_{14}Cl_2N_2O$ requires: 297, found: 298 $[M+H]^+$. ^1H NMR (500 MHz, DMSO-d_6) δ 7.96 (d, J = 5.5 Hz, 1H), 7.25 (t, J = 31.7 Hz, 2H), 6.86 (d, J = 8.7 Hz, 3H), 4.76 (s, 2H), 4.60 (d, J = 15.5 Hz, 2H), 3.77 – 3.68 (m, 3H).

Step 4: 6-((3-chloro-2-(4-methoxybenzylamino)pyridin-4-yl)methoxy)-N-cyclohexylbenzo[d]thiazol-2-amine. A mixture of 3-chloro-4-(chloromethyl)-N-(4-methoxybenzyl)pyridin-2-amine (51 mg, 0.17 mmol), 2-(cyclohexylamino)benzo[d]thiazol-6-ol (42 mg, 0.17 mmol) and Cs_2CO_3 (110 mg, 0.338 mmol) in DMF (2 mL) was stirred at 80°C for 3 h. The volatiles were removed under reduced pressure. The residue was purified by preparative HPLC (Mobile phase: A = 0.1% TFA/H_2O, B = 0.1% TFA/MeCN; Gradient: B = 5 - 95%; 12 min; Column: C18) to provide 6-((3-chloro-2-(4-methoxybenzylamino)pyridin-4-yl)methoxy)-N-cyclohexylbenzo[d]thiazol-2-amine (31 mg, 35%) as a white solid. MS (ES+) $C_{27}H_{29}ClN_4O_3S$ requires: 524, found: 525 $[M+H]^+$.

Step 5: 6-((2-amino-3-chloropyridin-4-yl)methoxy)-N-cyclohexylbenzo[d]thiazol-2-amine. A mixture of 6-((3-chloro-2-(4-methoxybenzylamino)pyridin-4-yl)methoxy)-N-

cyclohexylbenzo[d]thiazol-2-amine (31 mg, 0.057 mmol) in TFA (2 mL) was stirred at rt for 8 h. The volatiles were removed under reduced pressure. The residue was purified by preparative HPLC (Mobile phase: A = 0.1% NH_4HCO_3/H_2O, B = MeCN; Gradient: B = 5 - 95%; 12 min; Column: C18) to provide 6-((2-amino-3-chloropyridin-4-yl)methoxy)-N-cyclohexylbenzo[d]thiazol-2-amine (7 mg, 18%) as a white solid. MS (ES+) $C_{19}H_{21}ClN_4OS$ requires: 388, found: 389 [M+H]+. ^{1}H NMR (400 MHz, DMSO-d_6) δ 8.66 (s, 1H), 7.97 (d, J = 5.7 Hz, 1H), 7.49 (d, *J* = 2.5 Hz, 1H), 7.36 (d, *J* = 8.8 Hz, 1H), 7.00 (d, *J* = 8.8 Hz, 1H), 6.86 (d, *J* = 5.7 Hz, 1H), 5.18 (s, 2H), 3.68 (dt, *J* = 13.5, 8.7 Hz, 1H), 2.06 – 1.91 (m, 2H), 1.80 – 1.65 (m, 2H), 1.58 (dd, *J* = 16.3, 10.0 Hz, 1H), 1.29 (qd, *J* = 25.5, 12.8 Hz, 5H).

Synthesis of (1S,2S)-2-(6-((2-amino-3-chloropyridin-4-yl)methoxy)benzo[d]thiazol-2-ylamino)cyclohexanol (11)

Step 1: (1S,2S)-2-((6-methoxybenzo[d]thiazol-2-yl)amino)cyclohexanol. To a solution of 2-bromo-6-methoxybenzo[d]thiazole (200 mg, 0.82 mmol) and (1S,2S)-2-aminocyclohexanol (145mg, 1.23 mmol) in DMA (1 mL) was added DIPEA (0.28 mL,

1.64 mmol) and the resulting mixture was stirred at 100°C for 12 h. The reaction mixture was diluted with EtOAc (15 mL) and washed with water (10 mL). The layers were separated, and the organic layer was washed with brine (10 mL), dried over $MgSO_4$, filtered and concentrated under reduced pressure. The residue was purified via silica gel chromatography (40 to 80 % EtOAc in hexanes to provide (1S,2S)-2-((6-methoxybenzo[d]thiazol-2-yl)amino)cyclohexanol (180 mg, 0.647 mmol, 79 % yield) as a white solid. MS (ES+) $C_{14}H_{18}N_2O_2S$ requires: 278, found: 279 [M+H] +.

Step 2: 2-(((1S,2S)-2-hydroxycyclohexyl)amino)benzo[d]thiazol-6-ol. To a solution of (1S,2S)-2-((6-methoxybenzo[d]thiazol-2-yl)amino)cyclohexanol (150mg, 0.54 mmol) in DCM (5 mL) at 0 °C, was added BBr_3 (51 µL, 0.54mmol) slowly. The resulting mixture was stirred at rt for 3 h. The reaction mixture was then quenched slowly with ice-water (5 mL) followed by sat. $NaHCO_3$ (5 mL).The reaction mixture was diluted with EtOAc (20 ml).The layers were separated, and the organic layer was washed with $NaHCO_3$ (5 mL), dried over $MgSO_4$, filtered and concentrated under reduced pressure to give 2-(((1S,2S)-2-hydroxycyclohexyl)amino)benzo[d]thiazol-6-ol (120 mg, 84% yield) as white powder. MS (ES+) $C_{13}H_{16}N_2O_2S$ requires: 264, found: 265 [M+H] +.

Step 3: (1S,2S)-2-(6-((3-chloro-2-(4-methoxybenzylamino)pyridin-4-yl)methoxy)benzo[d]thiazol-2-ylamino)cyclohexanol. A mixture of 3-chloro-4-(chloromethyl)-N-(4-methoxybenzyl)pyridin-2-amine (50 mg, 0.17 mmol), 2-((1S,2S)-2-hydroxycyclohexylamino)benzo[d]thiazol-6-ol (45 mg, 0.17 mmol) and Cs_2CO_3 (110 mg, 0.34 mmol) in DMF (2 mL) was stirred at 80°C for 3 h. The volatiles were removed under reduced pressure. The residue was purified by preparative HPLC (Mobile phase: A = 0.1% TFA/H_2O, B = 0.1% TFA/MeCN; Gradient: B = 5 - 95%; 12 min; Column: C18) to give the title compound (30 mg, 34%) as a white solid. MS (ES+) $C_{27}H_{29}ClN_4O_3S$ requires: 524, found: 525 $[M+H]^+$.

Step 4: (1S,2S)-2-(6-((2-amino-3-chloropyridin-4-yl)methoxy)benzo[d]thiazol-2-ylamino)cyclohexanol. A mixture of (1S,2S)-2-(6-((3-chloro-2-(4-methoxybenzylamino)pyridin-4-yl)methoxy)benzo[d]thiazol-2-ylamino)cyclohexanol (30 mg, 0.06 mmol) in TFA (2 mL) was stirred at rt for 8 h. The volatiles were removed under reduced pressure. The residue was purified by preparative HPLC (Mobile phase: A = 0.1% NH_4HCO_3/H_2O, B = MeCN; Gradient: B = 5 - 95%; 12 min; Column: C18) to provide the title compound (17 mg, 73%) as a white solid. MS (ES+) $C_{19}H_{21}ClN_4O_2S$ requires:

404, found: 405 $[M+H]^+$. ^1H NMR (400 MHz, DMSO-d_6) δ 7.90 (d, J = 5.0 Hz, 1H), 7.74 (d, J = 7.5 Hz, 1H), 7.37 (d, J = 2.6 Hz, 1H), 7.27 (d, J = 8.7 Hz, 1H), 6.88 (dd, J = 8.7, 2.6 Hz, 1H), 6.72 (d, J = 5.0 Hz, 1H), 6.34 (s, 2H), 5.07 (s, 2H), 4.75 (d, J = 5.1 Hz, 1H), 3.56 – 3.47 (m, 1H), 3.41 – 3.35 (m, 1H), 2.07 (dd, J = 13.4, 7.7 Hz, 1H), 1.93 – 1.85 (m, 1H), 1.68 – 1.57 (m, 2H), 1.32 – 1.14 (m, 4H).

Synthesis of (1R,2R)-2-((6-((2-Amino-3-chloropyridin-4-yl)methoxy)benzo[d]thiazol-2-yl)amino)cyclohexan-1-ol (12)

Step 1: (1R,2R)-2-((6-Methoxybenzo[d]thiazol-2-yl)amino)cyclohexan-1-ol. A mixture of 2-bromo-6-methoxybenzo[d]thiazole (3.00 g, 12.3 mmol) and (1R,2R)-2-aminocyclohexanol (4.25 g, 36.9 mmol) was heated at 110 °C for 6 h. The reaction was cooled to rt, diluted with water (30 mL), and extracted with EtOAc (3 x 30 mL). The combined organic extracts were dried over Na_2SO_4 and concentrated to provide (1R,2R)-2-((6-methoxybenzo[d]thiazol-2-yl)amino)cyclohexan-1-ol (3.01 g, 88%) as a brown solid. MS (ES+) $C_{14}H_{18}N_2O_2S$ requires: 278, found: 279 $[M+H]^+$. ^1H NMR (500 MHz, $CDCl_3$) δ 7.37 (d, J = 11 Hz, 1H), 7.03 (d, J = 3.5 Hz, 1H), 6.84 (dd, J = 11, 3.5 Hz, 1H), 6.27 (bs,

1H), 3.78 (s, 3H), 3.4-3.35 (m, 2H), 2.12-2.01 (m, 2H), 1.70-1.65 (m, 2H), 1.38-1.16 (m, 4H).

Step 2: 2-(((1R,2R)-2-Hydroxycyclohexyl)amino)benzo[d]thiazol-6-ol. To a solution of 2-(6-methoxybenzo[d]thiazol-2-ylamino)cyclohexanol (3.00 g, 10.8 mmol) in DCM (30 mL) at 0 °C, was added boron tribromide (5.4 g, 21 mmol) slowly. The resulting mixture was stirred at rt for 3 h. The reaction mixture was then diluted slowly with ice-water (20 mL) followed by sat. $NaHCO_3$ (10 mL). The resulting precipitate was filtered and collected to provide 2-(((1R,2R)-2-hydroxycyclohexyl)amino)benzo[d]thiazol-6-ol (2.6 g, 90%) as a tan solid. MS (ES+) $C_{13}H_{16}N_2O_2S$ requires: 264, found: 265 $[M+H]^+$. ^1H NMR (500 MHz, d_4-MeOD) δ 7.12 (d, J = 9 Hz, 1H), 6.89 (d, J = 2 Hz, 1H), 6.62 (dd, J = 9, 2 Hz, 1H), 3.50-3.41 (m, 1H), 3.31-3.36 (m, 1H), 2.10-2.03 (m, 1H), 1.98-1.93 (m, 1H), 1.70-1.59 (m, 2H), 1.36-1.17 (m, 4H).

Step 3: (1R,2R)-2-(6-((3-Chloro-2-(4-methoxybenzylamino)pyridin-4-yl)methoxy) benzo[d]thiazol-2-ylamino)cyclohexanol. A mixture of 3-chloro-4-(chloromethyl)-N-(4-methoxybenzyl)pyridin-2-amine (50 mg, 0.17 mmol), 2-((1R,2R)-2-hydroxycyclohexylamino)benzo[d]thiazol-6-ol (45 mg, 0.17 mmol) and Cs_2CO_3 (110 mg,

0.34 mmol) in DMF (2 mL) was stirred at 80 °C for 3 h. The volatiles were removed under reduced pressure. The residue was purified by preparative HPLC (Mobile phase: A = 0.1% TFA/H_2O, B = 0.1% TFA/MeCN; Gradient: B = 5 - 95%; 12 min; Column: C18) to provide (1R,2R)-2-(6-((3-chloro-2-(4-methoxybenzylamino)pyridin-4-yl)methoxy) benzo[d]thiazol-2-ylamino)cyclohexanol (30 mg, 34%) as a white solid. MS (ES+) $C_{27}H_{29}ClN_4O_3S$ requires: 524, found: 525 $[M+H]^+$.

Step 4: (1R,2R)-2-(6-((2-Amino-3-chloropyridin-4-yl)methoxy)benzo[d]thiazol-2-ylamino)cyclohexanol. A mixture of (1R,2R)-2-(6-((3-chloro-2-(4-methoxybenzylamino)pyridin-4-yl)methoxy) benzo[d]thiazol-2-ylamino)cyclohexanol (30 mg, 0.06 mmol) in TFA (2 mL) was stirred at RT for 8 h. The volatiles were removed under reduced pressure. The residue was purified by preparative HPLC (Mobile phase: A = 0.1% NH_4HCO_3/H_2O, B = MeCN; Gradient: B = 5 - 95%; 12 min; Column: C18) to give (1R,2R)-2-(6-((2-amino-3-chloropyridin-4-yl)methoxy)benzo[d]thiazol-2-ylamino)cyclohexanol (5 mg, 21%) as a white solid. HRMS (ES+) $C_{19}H_{22}ClN_4O_2S^+$ requires: 405.1147, found: 405.1151 $[M+H]^+$. ^1H NMR (500 MHz, *d_6*- DMSO-*d_6*) δ 7.90 (d, *J* = 5.0 Hz, 1H), 7.72 (d, *J* = 7.5 Hz, 1H), 7.36 (d, *J* = 2.5 Hz, 1H), 7.26 (d, *J* = 8.5 Hz, 1H),

6.88 (dd, *J* = 8.5, 2.5 Hz, 1H), 6.72 (d, *J* = 5.0 Hz, 1H), 6.31 (s, 2H), 5.07 (s, 2H), 4.73 (d, *J* = 5.5 Hz, 1H), 3.52~3.48 (m, 1H), 3.36~3.31 (m, 1H), 2.06~2.04 (m, 1H), 1.89~1.86 (m, 1H), 1.64 – 1.60 (m, 2H), 1.29 – 1.17 (m, 4H). ^{13}C NMR (600 MHz, DMSO-d_6) δ 165.00, 155.76, 152.61, 147.29, 145.97, 143.84, 131.20, 117.99, 113.45, 111.59, 111.18, 106.80, 71.38, 66.70, 59.48, 39.89, 39.75, 39.61, 39.47, 39.33, 39.19, 39.05, 33.98, 30.64, 23.95, 23.60.

Synthesis of (1R,2S)-2-((6-((2-amino-3-chloropyridin-4-yl)methoxy)benzo[d]thiazol-2-yl)amino)cyclohexanol (13)

Step 1: 2-chlorobenzo[d]thiazol-6-ol. To a solution of 2-chloro-6-methoxybenzo[d]thiazole (150 g, 751 mmol, 1.0 *eq*) in DCM (1.5 L) was added BBr_3 (470 g, 1.9 mol, 180 mL, 2.5 *eq*) drop-wise at -10°C - 0°C. During the addition, the solid was precipitated. After the addition, the reaction mixture was stirred at 25°C for 16 hr. Three additional batches of the same reaction were run. The four batches were combined and the mixture was poured into cooled water (10 L) slowly, then $NaHCO_3$ (sat.) solution (6 L) was added. The mixture was stirred at 0°C - 5°C for 0.5h, filtered and the filter cake was washed with water (~10 L) until the pH of filtrate was 7. The filter cake was washed

with pet. ether (5 L) and dried under vacuum to afford 2-chlorobenzo[d]thiazol-6-ol (507 g, 2.59 mol, 86% yield) as the white solid. The product was used in the next step without further purification. MS (ES+) C_7H_4ClNOS requires: 185, found: 186 $[M+H]^+$.

Step 2: 3-chloro-4-(((2-chlorobenzo[d]thiazol-6-yl)oxy)methyl)-N-(4-methoxybenzyl)pyridin-2-amine. To a solution of 2-chlorobenzo[d]thiazol-6-ol (143g, 733 mmol) in DMF (800 mL) was added 3-chloro-4-(chloromethyl)-N-(4-methoxybenzyl)pyridin-2-amine (220 g, 733 mmol) and Cs_2CO_3 (477 g, 1.47) at 15°C. The mixture was stirred at 25°C for 16 hr. Water (1L) was added drop-wise and the solid was precipitated. The suspension was stirred at 25°C for 1 h. Three additional batches of the same reaction were run. The three batches were combined, filtered, washed with water (10 L) and the filter cake was collected. The cake was sub packaged slurried in pet ether/EtOAc = 5/1 (2L, two times), filtered, collected and dried under vacuum to provide 3-chloro-4-(((2-chlorobenzo[d]thiazol-6-yl)oxy)methyl)-N-(4-methoxybenzyl)pyridin-2-amine (800g, 1.76 mol, 80%) as the light yellow solid. MS (ES+) $C_{21}H_{17}Cl_2N_3O_2S$ requires: 445, found: 446 $[M+H]^+$.

Step 3: (1R,2S)-2-((6-((2-amino-3-chloropyridin-4-yl)methoxy)benzo[d]thiazol-2-yl)amino)cyclohexanol. To a solution of 3-chloro-4-(((2-chlorobenzo[d]thiazol-6-yl)oxy)methyl)-N-(4-methoxybenzyl)pyridin-2-amine (2.0 g, 4.5 mmol) in NMP (20 mL) was added Na_2CO_3 (2.4 g, 22 mmol) and (1R,2S)-2-aminocyclohexan-1-ol (0.81 mg, 5.4 mmol,). The mixture was stirred at 125°C for 16 hr. The mixture was poured into water (100 mL), extracted with ethyl acetate (2 x 100 mL) and the residue was purified by pre-HPLC (Mobile phase: A = 0.1% NH_4HCO_3/H_2O, B = MeCN; Gradient: B = 5 - 95%; 12 min; Column: C18) to provide the (1R,2S)-2-((6-((3-chloro-2-((4-methoxybenzyl)amino)pyridin-4-yl)methoxy)benzo[d]thiazol-2-yl)amino)cyclohexan-1-ol (1.58 g, 2.98 mmol, 66% yield) as a brown solid. ^{1}H NMR (400 MHz, DMSO-d_6) δ: 7.94 (d, J = 4.8 Hz, 1H), 7.62 (d, J = 8.0 Hz, 1H), 7.37 (d, J = 2.4 Hz, 1H), 7.23 - 7.28 (m,3H), 7.09 (t, J = 6.4 Hz, 1H), 6.84 - 6.88 (m, 3H), 6.72 (d, J = 4.8 Hz, 1H), 5.08 (s, 2H), 4.65 (d, J = 4.0 Hz, 1H), 4.53 (d, J = 6.4 Hz, 2H), 3.92 (s, 1H), 3.79 - 3.84 (m, 1H), 3.71 (s, 3H),1.48 - 1.71 (m, 6H), 1.29 - 1.34 (m, 2H).

Step 4: (1R,2S)-2-((6-((2-amino-3-chloropyridin-4-yl)methoxy)benzo[d]thiazol-2-yl)amino)cyclohexanol: A solution of (1R,2S)-2-((6-((3-chloro-2-((4-

methoxybenzyl)amino)pyridin-4-yl)methoxy)benzo[d]thiazol-2-yl)amino)cyclohexan-1-ol (1.0 g, 1.9 mmol) in TFA (10 mL) was stirred at 25°C for 16 hr. The mixture was quenched with water (100 mL) and the pH was adjusted to 9 with NH_4OH (aq., sat.). The solids were filtered and collected. The solid was purified by slurrying with EtOAc and Pet. Ether 1:3 (40 mL). The mixture was filtered to collect the solids and provide the product (500 mg, 1.22 mmol, 64% yield) as a light brown solid. ^{1}H NMR (400 MHz, DMSO-d_6) δ: 7.90 (d, J = 4.8 Hz, 1H), 7.61 (d, J = 7.6 Hz, 1H), 7.36 (d, J = 2.4 Hz, 1H), 7.27 (d, J = 8.8 Hz, 1H), 6.87 (dd, J = 2.4 Hz, 8.4 Hz, 1H), 6.72 (d, J = 5.2 Hz, 1H), 6.31 (s, 2H), 5.07 (s, 2H), 4.64 (d, J = 4.0 Hz, 1H), 3.92 (s, 1H), 3.79 - 3.84 (m, 1H), 1.48 - 1.71 (m, 6H), 1.29 - 1.34 (m, 2H).

Synthesis of (1S,2R)-2-((6-((2-amino-3-chloropyridin-4-yl)methoxy)benzo[d]thiazol-2-yl)amino)cyclohexanol (14)

Step 1: Synthesis of (1S,2R)-2-((6-((2-amino-3-chloropyridin-4-yl)methoxy)benzo[d]thiazol-2-yl)amino)cyclohexanol. To a solution of 3-chloro-4-(((2-chlorobenzo[d]thiazol-6-yl)oxy)methyl)-N-(4-methoxybenzyl)pyridin-2-amine (2.0 g, 3.8 mmol) in NMP (10 mL) were added (1S,2R)-2-aminocyclohexanol (685 mg, 4.52 mmol) and Na_2CO_3 (2.0 g,19 mmol). The reaction mixture was stirred at 120°C for 16 hr. Water was added (100 mL), and the mixture was extracted with EtOAc (3 x 100 mL). The organic

phases were washed with brine (2 x 100 mL), dried over Na_2SO_4, filtered and concentrated under reduced pressure. The residue was purified by prep-HPLC (Mobile phase: A = 0.1% NH_4HCO_3/H_2O, B = MeCN; Gradient: B = 5 - 95%; 12 min; Column: C18) to provide (1S,2R)-2-((6-((3-chloro-2-((4-methoxybenzyl)amino)pyridin-4-yl)methoxy)benzo[d]thiazol-2-yl)amino)cyclohexan-1-ol (2.9 g, 5.3 mmol, 70% yield) as the gray solid. ^{1}H NMR (400 MHz, DMSO-d_6) δ: 7.94 (d, J = 5.2 Hz, 1H), 7.61 (d, J = 7.6 Hz, 1H), 7.36 (d, J = 2.8 Hz, 1H), 7.27 - 7.23 (m, 3H), 7.07(t, J = 6.0 Hz, 1H), 6.87 - 6.83 (m, 3H), 6.71 (d, J = 5.2 Hz, 1H), 5.07 (s, 2H), 4.64 (d, J =4.0 Hz, 1H), 4.53 (d, J = 6.0 Hz, 2H), 3.91 (s, 1H),3.81 (t, J = 8.0 Hz, 1H), 3.70(s, 3H), 1.70 - 1.44 (m, 6H), 1.33 - 1.28 (m, 2H).

Step 2: (1S,2R)-2-((6-((2-amino-3-chloropyridin-4-yl)methoxy)benzo[d]thiazol-2-yl)amino)cyclohexanol. (1S,2R)-2-((6-((3-Chloro-2-((4-methoxybenzyl)amino)pyridin-4-yl)methoxy)benzo[d]thiazol-2-yl)amino)cyclohexan-1-ol (2.3 g, 4.2 mmol) in TFA (23 mL) was stirred at rt for 16 h. Water (30 ml) was added and the pH was adjusted to 9 with NH_4OH (aq., sat.). The solids were filtered and collected. The solid was purified by slurrying with EtOAc and Pet. Ether 1:3 (40 mL). The mixture was filtered and the solids were collected to provide

the product (804 mg, 1.92 mmol, 45%) as a yellow solid. ^{1}H NMR (400 MHz, DMSO-d_6) δ: 7.89 (d, J = 5.2 Hz, 1H), 7.60 (d, J = 8.0 Hz, 1H), 7.36 (d, J = 2.8 Hz, 1H), 7.27 (d, J = 8.4Hz, 1H), 6.87 (dd, J = 2.4, 8.8 Hz, 1H), 6.72 (d, J = 4.8 Hz, 1H), 6.31 (s, 2H), 5.06 (s, 2H), 4.64 (d, J = 4.0 Hz, 1H), 3.91 (s, 1H), 3.83 - 3.78 (m, 1H), 1.70 - 1.44 (m, 6H), 1.31 - 1.28 (m, 2H).

Synthesis of (1R,2R)-2-({6-[(2-Amino-3-chloropyridin-4-yl)methoxy]-4-methoxy-1,3-benzothiazol-2-yl}amino)cyclohexan-1-ol (15)

Step 1: 4-(benzyloxy)-2-methoxy-1-nitrobenzene. To a solution of phenylmethanol (41.0 g, 380 mmol) in DMF (500 ml) was added NaH (9.60 g, 399 mmol) at 0^{o}C. The reaction mixture was stirred for 0.5 h then 4-fluoro-2-methoxy-1-nitrobenzene (50.0 g, 292 mmol) was added. The mixture was stirred at RT for 1 h. The reaction was quenched with water (500 mL), filtered and concentrated under reduced pressure to give 4-(benzyloxy)-2-methoxy-1-nitrobenzene (75 g, 99%) as yellow solid. LC-MS (ES+) $C_{14}H_{13}NO_4$ requires: 259, found: 260 $(M+H)^+$.

Step 2: 4-(Benzyloxy)-2-methoxybenzenamine. To a solution of 4-(benzyloxy)-2-methoxy-1-nitrobenzene (75.0 g, 290 mmol) in MeOH (800 mL) was added Raney-Ni (3.0

g) followed by the dropwise addition of $N_2H_4.H_2O$ (44.0 g, 870 mmol) at 0 °C. the mixture was stirred at rt for 16h. The reaction mixture was quenched by sat. NH_4OH (200 mL) and extracted with EtOAc (2 x 100 mL). The organic layers were dried, filtered and concentrated under reduced pressure to give 4-(benzyloxy)-2-methoxybenzenamine (56 g, 84 %) as yellow liquid. LC-MS (ES+) $C_{14}H_{15}NO_2$ requires: 229, found: 230 $(M+H)^+$.

Step 3: 6-(Benzyloxy)-4-methoxybenzo[d]thiazol-2-amine. To a solution of 4-(benzyloxy)-2-methoxybenzenamine (28.0 g, 0.122 mol) in methanol (600 mL) was added KSCN (59.3g, 0.611 mol), followed by the addition of anhydrous $CuSO_4$ (195g, 1.22 mol). The mixture was stirred for 16h at 90 °C and concentrated under reduced pressure. The residue was dissolved in DCM (500 mL), the precipitate was filtered and the filter cake was washed with dichloromethane (2 x 100 mL). The combined organic layer was washed with NH_4OH (aq., sat., 500 mL). The aqueous layer was extracted with dichloromethane. The combined organic layers were concentrated under reduced pressure. The residue was purified by silica gel chromatography (50% to 100% EtOAc in pet. ether) to give 6-(benzyloxy)-4-methoxybenzo[d]thiazol-2-amine (13 g, 37%) as a black solid. LC-MS (ES+) $C_{15}H_{14}N_2O_2S$ requires: 286, found: 287 $(M+H)^+$.

Step 4: 6-(Benzyloxy)-2-bromo-4-methoxybenzo[d]thiazole. To a suspension of 6-(benzyloxy)-4-methoxybenzo[d]thiazol-2-amine (22.0 g, 0.077 mol) in MeCN (100 mL) was added t-BuONO (11.8 g, 0.110 mol) at 0 °C. The mixture was stirred for 30 min, then $CuBr_2$ (10.3 g, 0.046 mol) was added. The reaction was stirred for an additional 2 h at room temperature. Water was added, the precipitate was filtered and the filtrate was extracted with EtOAc. The combined organic layers were washed with water, diluted NH_4OH (aq., sat.), brine and concentrated under reduced pressure. The residue was purified by silica gel chromatography (10% EtOAc in Petroleum ether) to give 6-(benzyloxy)-2-bromo-4-methoxybenzo[d]thiazole (8.0 g, 30%) as a grey-white solid. LC-MS (ES+) $C_{15}H_{12}BrNO_2S$ requires: 349, found: 350 $(M+H)^+$. $(M+H)^+$. ^1H NMR (500 MHz, $CDCl_3$) δ 7.51 – 7.29 (m, 5H), 6.88 (d, *J* = 2.2 Hz, 1H), 6.61 (d, *J* = 2.2 Hz, 1H), 5.09 (s, 2H), 3.98 (s, 3H).

Step 5: (1R,2R)-2-(6-(Benzyloxy)-4-methoxybenzo[d]thiazol-2-ylamino)cyclohexanol. A mixture of 6-(benzyloxy)-2-bromo-4-methoxybenzo[d]thiazole (3.50 g, 10.0 mmol), (1R, 2R)-2-aminocyclohexanol (3.45 g, 30.0 mmol) and DIPEA (5.16 g, 40.0 mmol) in DMF (10 mmol) was heated at 110 °C for 48 h. The reaction was cooled to rt, diluted with water

(30 mL), and extracted with EtOAc (3 x 30 mL). The combined organic extracts were washed by water (100 ml), brine (100 ml), dried over Na_2SO_4 and concentrated to give (1R,2R)-2-(6-(benzyloxy)-4-methoxybenzo[d]thiazol-2-ylamino)cyclohexanol (3.8 g, 100%, crude) as a brown solid. MS (ES+) $C_{21}H_{24}N_2O_3S$ requires: 384, found: 385 $[M+H]^+$.

Step 6: 2-((1R,2R)-2-Hydroxycyclohexylamino)-4-methoxybenzo[d]thiazol-6-ol. A solution of (1R,2R)-2-(6-(benzyloxy)-4-methoxybenzo[d]thiazol-2-ylamino)cyclohexanol (1152 mg, 3.0 mmol) in TFA (10 mL) was heated at 65 °C for 48 h. The solvent was removed, the reaction mixture was diluted slowly by sat. $NaHCO_3$ and extracted with EtOAc (3 x 50 mL). The combined organic extracts were washed with water (100 ml) and brine (100 ml), dried over Na_2SO_4 and concentrated to give 2-((1R,2R)-2-hydroxycyclohexylamino)-4-methoxybenzo[d]thiazol-6-ol (882 mg, 100%, crude) as a brown solid. MS (ES+) $C_{14}H_{18}N_2O_3S$ requires: 294, found: 295 $[M+H]^+$.

Step 7: 2-((1R,2R)-2-Hydroxycyclohexylamino)-4-methoxybenzo[d]thiazol-6-ol. A mixture of 3-chloro-4-(chloromethyl)-N-(4-methoxybenzyl)pyridin-2-amine (Example 1, Step 3; 202 mg, 0.68 mmol), 2-((1R,2R)-2-hydroxycyclohexylamino)-4-

methoxybenzo[d]thiazol-6-ol (200 mg, 0.68 mmol) and Cs_2CO_3 (443 mg, 1.36 mmol) in DMF (2 mL) was stirred at 80 ℃ for 3 h. The mixture was diluted by water (30 ml) and extracted with EtOAc (3 × 30 mL). The combined organic phases were washed with water (100 ml) and brine (100 ml), dried over Na_2SO_4 and concentrated to give (1R,2R)-2-(6-((3-chloro-2-(4-methoxybenzylamino)pyridin-4-yl)methoxy)-4-methoxybenzo[d]thiazol-2-ylamino)cyclohexanol (300 mg, 79%, crude) as a brown solid. MS (ES+) $C_{28}H_{31}ClN_4O_4S$ requires: 554, found: 555 $[M+H]^+$.

Step 8: (1R,2R)-2-(6-((2-Amino-3-chloropyridin-4-yl)methoxy)-4-methoxybenzo[d]thiazol-2-ylamino)cyclohexanol. A mixture of (1R,2R)-2-(6-((3-chloro-2-(4-methoxybenzylamino)pyridin-4-yl)methoxy)-4-methoxybenzo[d]thiazol-2-ylamino)cyclohexanol (300 mg, 0.54 mmol) in TFA (2 mL) was stirred at rt for 8 h. The volatiles were removed under reduced pressure. The residue was purified by preparative HPLC (Mobile phase: A = 0.1% NH_4HCO_3/H_2O, B = MeCN; Gradient: B = 5 - 95%; 12 min; Column: C18) to give (1R,2R)-2-(6-((2-amino-3-chloropyridin-4-yl)methoxy)-4-methoxybenzo[d]thiazol-2-ylamino)cyclohexanol (19 mg, 8%) as a white solid. MS (ES+) $C_{20}H_{23}ClN_4O_3S$ requires: 434, found: 435 $[M+H]^+$. ^1H NMR (500 MHz, DMSO-d_6) δ 8.50

(s, 1H), 7.95 (d, *J* = 5.4 Hz, 1H), 7.04 (s, 1H), 6.85 (d, *J* = 5.5 Hz, 1H), 6.69 (s, 1H), 5.14 (s, 2H), 3.88 (s, 3H), 3.60 (d, *J* = 11.0 Hz, 1H), 3.32 (dd, *J* = 17.5, 11.8 Hz, 1H), 2.01 (d, *J* = 13.0 Hz, 1H), 1.89 (d, *J* = 8.9 Hz, 1H), 1.70 – 1.56 (m, 2H), 1.42 – 0.92 (m, 4H).

(1R,2R)-2-(6-((2-Amino-3-chloropyridin-4-yl)methoxy)-4-(trifluoromethoxy)benzo[d]thiazol-2-ylamino)cyclohexanol (16)

Step 1: 6-Bromo-4-(trifluoromethoxy)benzo[d]thiazol-2-amine. To a solution of 4-bromo-2-(trifluoromethoxy)aniline (5.9 g, 23.1 mmol) in AcOH (100 ml) was added KSCN (8.97 g, 92.5 mmol). After stirring for 30 min at rt, the reaction mixture was cooled to 0 ^{o}C, and a solution of Br_2 (5.5 g, 34.6 mmol) in AcOH was added dropwise. The reaction mixture was stirred for 4 h at rt. Subsequently, the pH of the reaction mixture was adjusted to 7 with NH_3-H_2O. The mixture was extracted with EtOAc (3 × 80 mL), the combined organic phases were dried over Na_2SO_4 and concentrated under reduced pressure. The residue was purified by column chromatography (PE: THF = 10:1) to give the title compound (0.9 g, 12.5%) MS (ES+) $C_8H_4BrF_3N_2OS$ requires: 312, found: 313 $[M+H]^+$.

Step 2: 2,6-Dibromo-4-(trifluoromethoxy)benzo[d]thiazole. A solution of 6-bromo-4-(trifluoromethoxy)benzo[d]thiazol-2-amine (900 mg, 3 mmol) in MeCN (20 mL) was cooled to -5^{o}C and $CuBr_2$ (0.83 g, 3.6 mmol) and ^{t}BuONO (0.37 mg, 3.6 mmol) were added dropwise. The reaction mixture was stirred at 0-5^{o}C for 30 min, then it was heated to 40^{o}C and stirred for 6 h. The insoluble was filtered off and the filtrate was washed with 1 N HCl solution, the resultant solution was dried over Na_2SO_4, filtered and concentrated to give the product (0.6 g, 53%). MS (ESI+) $C_8H_2Br_2F_3NOS$ requires: 375, found: 376 $[M+H]^+$.

Step 3: (1R,2R)-2-(6-Bromo-4-(trifluoromethoxy)benzo[d]thiazol-2-ylamino)cyclohexanol.

A mixture of 2,6-dibromo-4-(trifluoromethoxy)benzo[d]thiazole (980 mg, 2.61 mmol), (1R,2R)-2-aminocyclohexanol (901 mg, 7.83 mmol) and DIPEA (1 g, 7.83 mmol) in DMA (10 ml) was heated to 100^{o}C for 16 h. Saturated NH_4Cl solution (8 ml) was added and the reaction mixture was extracted with EtOAc (3 x 5 mL). The combined organic phases were dried over Na_2SO_4, filtered and concentrated. The residue was purified by column

chromatography (25% EtOAc in pet. ether) to give the product (836 mg, 78%) as a white solid. MS (ESI+) $C_{14}H_{14}BrF_3N_2O_2S$ requires: 410, found: 411 $[M+H]^+$.

Step 4: (1R,2R)-2-(6-(4,4,5,5-Tetramethyl-1,3,2-dioxaborolan-2-yl)-4-(trifluoromethoxy)benzo[d]thiazol-2-ylamino)cyclohexanol. A mixture of (1R, 2R)-2-(6-bromo-4-(trifluoromethoxy)benzo[d]thiazol-2-ylamino)cyclohexanol (600 mg, 1.463 mmol), B_2Pin_2 (446 mg, 1.75 mmol), KOAc (286 mg, 2.92 mmol), $Pd(dppf)Cl_2$ (107 mg, 0.15 mmol) in 1,4-dioxane (20 ml) was heated to 100 ^oC and stirred for 16h. The mixture was filtered and the filtrate was concentrated under reduced pressure to provide the residue that was used in the next step without further purification (671 mg, 100%). MS (ESI+) $C_{20}H_{26}BF_3N_2O_4S$ requires: 458, found: 459 $[M+H]^+$.

Step 5: 2-((1R,2R)-2-Hydroxycyclohexylamino)-4-(trifluoromethoxy)benzo[d]thiazol-6-ol. To a solution of (1R,2R)-2-(6-(4,4,5,5-tetramethyl-1,3,2-dioxaborolan-2-yl)-4-(trifluoromethoxy)benzo[d]thiazol-2-ylamino)cyclohexanol (700 mg, 1.53 mmol) in dioxane (5 ml) was added H_2O_2 (5 ml) and the reaction mixture was stirred for 2 h at rt. The solvent was evaporated and the residue was taken up in EtOAc (5 ml)/water (4 ml). The crude product was extracted with EtOAc (3 x 5 mL), the combined organic phases

were dried over Na_2SO_4, filtered and concentrated under reduced pressure. The residue was purified by column chromatography (25% EtOAc in pet. ether) to provide the product (320 mg, 60%) as an oil. MS (ESI+) $C_{14}H_{15}F_3N_2O_3S$ requires: 348, found: 349 $[M+H]^+$.

Step 6: (1R,2R)-2-(6-((3-Chloro-2-(4-methoxybenzylamino)pyridin-4-yl)methoxy)-4-(trifluoromethoxy)benzo[d]thiazol-2-ylamino)cyclohexanol. A mixture of 2-((1R,2R)-2-hydroxycyclohexylamino)-4-(trifluoromethoxy)benzo[d]thiazol-6-ol (150 mg, 0.43 mmol), 3-chloro-4-(chloromethyl)-N-(4-methoxybenzyl)pyridin-2-amine (153 mg, 0.517 mmol) and Cs_2CO_3 (421 mg, 1.29 mmol) in DMF (3 ml) was stirred at 80°C for 16 h. The volatiles were removed under reduced pressure and the residue was purified by column chromatography (70% EtOAc in pet. ether) to provide the product (70 mg, 27%) as an oil. MS (ESI+) $C_{28}H_{28}ClF_3N_4O_4S$ requires: 608, found: 609 $[M+H]^+$.

Step 7: (1R,2R)-2-(6-((2-Amino-3-chloropyridin-4-yl)methoxy)-4-(trifluoromethoxy)benzo[d]thiazol-2-ylamino)cyclohexanol. To a solution of (1R, 2R)-2-(6-((3-chloro-2-(4-methoxybenzylamino)pyridin-4-yl)methoxy)-4-(trifluoromethoxy)benzo[d]thiazol-2-ylamino)cyclohexanol (70 mg, 0.115 mmol) in DCM (3 mL) was added TFA (3 ml) , then the reaction mixture was stirred at rt for 16 h. The

solvent was evaporated and 2N NaOH was added until the pH was adjusted to > 7. The mixture was stirred for 1 h and the crude product was extracted with EtOAc (3 x 3 mL). The combined organic phases were dried over Na_2SO_4, filtered and concentrated under reduced pressure. The residue was purified by column chromatography (90% EtOAc in pet. ether) to give the title compound (2.5 mg, 4.5%) as a white solid. MS (ES+) $C_{20}H_{20}ClF_3N_4O_3S$ requires: 488, found: 489.5 $[M+H]^+$. ^1H NMR (500 MHz, DMSO-d_6) δ 8 8.14 (d, J = 6.5 Hz, 1H), 7.91 (d, J = 5 Hz, 1H), 7.49 (d, J = 2.5 Hz, 1H), 6.94 (s, 1H), 6.74 (d, J = 4.5 Hz, 1H), 6.35 (s, 2H), 5.11 (s, 2H), 4.80 (d, J = 4.5 Hz, 1H), 3.34-3.40 (m, 1H), 2.04-2.06 (m, 1H), 1.87-1.90 (m, 1H), 1.62-1.64 (m, 2H), 1.24-1.30 (m, 4H).

Synthesis of (1R,2R)-2-(6-((2-Amino-3-chloropyridin-4-yl)methoxy)-4-chlorobenzo[d]thiazol-2-ylamino)cyclohexanol (17)

Step 1: 4-Chloro-6-methoxybenzo[d]thiazol-2-amine. To a solution of 2-chloro-4-methoxyaniline (1.6 g, 10 mmol) in AcOH (20 ml) was added KSCN (3.95 g, 40.76 mmol) and $CuBr_2$ (2.6, 11.9 mmol) the reaction mixture was cooled to 0°C. Br_2 (1.95 g, 12.2 mmol) was added dropwise and the reaction mixture was stirred for 4 h at rt. The pH of the mixture was adjusted to 7 with NH_4OH (aq., sat.) solution. The mixture was extracted

with EtOAc (3 x 50 mL), the combined organic phases were dried over Na_2SO_4 and concentrated under reduced pressure. The residue was purified by column chromatography (30% EtOAc in pet. ether) to provide the 4-chloro-6-methoxybenzo[d]thiazol-2-amine (1.7 g, 78%) MS (ES+) $C_8H_7ClN_2OS$ requires: 214, found: 215 $[M+H]^+$. ^1H NMR (500 MHz, DMSO-d_6) δ 7.596 (s, 2H), 7.311(s, 1H), 6.925 (s, 1H), 3.749 (s, 3H).

Step 2: 2-Bromo-4-chloro-6-methoxybenzo[d]thiazole. A solution of 4-chloro-6-methoxybenzo[d]thiazol-2-amine (2.1 g, 7.9 mmol) in MeCN (40 mL) was cooled to -5°C and ^{t}BuONO (1.23g, 11.9 mmol) was added dropwise. The mixture was stirred at 0°C for 30 min, and subsequently heated to 40 °C and stirred for 6 hours. The mixture was filtered and the filtrate was washed with 1N HCl solution. The organic phase was dried over Na_2SO_4, filtered and concentrated under reduced pressure to provide the crude product (1.25 g, 46%) that was used in the next step without further purification. MS (ESI+) $C_8H_5BrClNOS$ requires: 277, found: 278 $[M+H]^+$.

Steps 3-7: (1R,2R)-2-(6-((2-Amino-3-chloropyridin-4-yl)methoxy)-4-chlorobenzo[d]thiazol-2-ylamino)cyclohexanol. As described for compound **12**. (1R,2R)-

2-(6-((2-Amino-3-chloropyridin-4-yl)methoxy)-4-chlorobenzo[d]thiazol-2-ylamino)cyclohexanol MS (ES+) $C_{19}H_{20}Cl_2N_4O_2S$ requires: 438, found: 439 $[M+H]^+$. ^1H NMR (500 MHz, DMSO-d_6) δ 8.06 (d, J = 8.0 Hz, 1H), 7.91 (d, J = 5 Hz, 1H), 7.40 (d, J = 2.5 Hz, 1H), 7.02 (d, J = 2.5 Hz, 1H), 6.72 (d, J = 5 Hz, 1H), 6.53 (s, 2H), 5.10 (s, 2H), 4.81 (d, J = 5 Hz, 1H), 3.36-3.52 (m, 1H), 1.99-2.02 (m, 1H), 1.87-1.89 (m, 1H), 1.62-1.64 (m, 2H), 1.15-1.31 (m, 4H).

Synthesis of (1R, 2R)-2-(6-((2-amino-3-chloropyridin-4-yl)methoxy)-4-fluorobenzo[d]thiazol-2-ylamino)cyclohexanol (18)

(1R, 2R)-2-(6-((2-Amino-3-chloropyridin-4-yl) methoxy)-4-fluorobenzo[d]thiazol-2-ylamino)cyclohexanol was prepared as described for compound 17. MS (ES+) $C_{19}H_{20}ClFN_4O_2S$ requires: 422, found: 423 $[M+H]^+$. ^1H NMR (500 MHz, DMSO-d_6) δ 7.957 (d, J = 7.5 Hz, 1H), 7.910 (d, J = 5 Hz, 1H), 7.251 (d, J = 2Hz, 1H), 7.864 (dd, J_1 = 2 Hz, J_2 = 12.5 Hz, 1H), 6.725 (d, J = 4.5 Hz, 1H), 6.350 (s, 1H), 5.087 (s, 2H), 4.761 (d, J = 5 Hz, 1H), 3.514-3.526 (m, 1H), 2.041-2.065 (m, 1H), 1.874-1.897 (m, 1H), 1.616-1.657 (m, 2H), 1.182-1.309 (m, 4H).

Synthesis of (1R,2R)-2-(6-((2-amino-3-chloropyridin-4-yl)methoxy)-7-chlorobenzo[d]thiazol-2-ylamino)cyclohexanol (19)

(1R,2R)-2-(6-((2-Amino-3-chloropyridin-4-yl)methoxy)-7-chlorobenzo[d]thiazol-2-ylamino)cyclohexanol was prepared as described for compound 17. MS (ES+) $C_{19}H_{20}Cl_2N_4O_2S$ requires: 438, found: 439 $[M+H]^+$. ^1H NMR (500 MHz, DMSO-d_6) δ 8.03 (d, J = 12.5 Hz, 1H), 7.93 (d, J = 5 Hz, 1H), 7.27 (d, J = 8.5 z, 1H), 7.07 (d, J = 8.5 Hz, 1H), 6.77 (d, J = 5 Hz, 1H), 6.37 (s, 2H), 5.18 (s, 2H), 4.77 (d, J = 5 Hz, 1H), 3.48-3.50 (m, 1H), 2.04-2.07 (m, 1H), 1.88-1.90 (m, 1H), 1.61-1.65 (m, 2H), 1.18-1.30 (m, 4H).

Synthesis of (1R,2R)-2-(6-((2-amino-3-chloropyridin-4-yl)methoxy)-7-fluorobenzo[d]thiazol-2-ylamino)cyclohexanol (20)

(1R,2R)-2-(6-((2-Amino-3-chloropyridin-4-yl)methoxy)-7-fluorobenzo[d]thiazol-2-ylamino)cyclohexanol was prepared as described for compound 17. MS (ES+) $C_{19}H_{20}ClFN_4O_2S$ requires: 422, found: 423.5 $[M+H]^+$. ^1H NMR (500 MHz, DMSO-d_6) δ 8.07 (d, J = 7.5 Hz, 1H), 7.91 (d, J = 5 Hz, 1H), 7.06-7.14 (m, 2H), 6.73 (d, J = 5 Hz, 1H), 6.36 (s, 2H), 5.16 (s, 2H), 4.77 (d, J = 5 Hz, 1H), 3.49-3.51 (m, 1H), 2.05-2.07 (m, 1H), 1.88-1.90 (m, 1H), 1.61-1.65 (m, 2H), 1.18-1.30 (m, 4H).

Synthesis of N-cyclohexyl-6-((pyridin-3-yloxy)methyl)benzo[d]thiazol-2-amine (4)

Step 1: (2-bromobenzo[d]thiazol-6-yl)methanol: To a solution ethyl 2-bromobenzo[d]thiazole-6-carboxylate (1.2 g, 4.2 mmol) in THF (20 mL), at 0°C, DIBAL-H (10.5 mL, 10.5 mmol, 1M in toluene) was added slowly. The reaction mixture was stirred at rt for 4 h. The mixture was quenched with water (15 ml), filtered and the filtrate was extracted with EtOAc (3 x 30 mL). The combined organic layers were dried over $MgSO_4$ and concentrated under reduced pressure to provide (2-bromobenzo[d]thiazol-6-yl)methanol (1.0 g, 98%) as a yellow solid. MS (ES+) C_8H_6BrNOS requires: 244, found: 244, 246$[M+H]^+$.

Step 2: (2-(methylthio)benzo[d]thiazol-6-yl)methanol. A mixture of (2-bromobenzo[d]thiazol-6-yl)methanol (1.0 g, 4.0 mmol) and CH_3SNa (560 mg, 8 mmol) in DMF (15 mL) was stirred at rt for 16h. The reaction mixture was diluted with water and extracted with EtOAs (3 x 30 mL). The combined organic layers were washed with brine, dried over $MgSO_4$ and concentrated under reduced pressure. The residue was purified by flash column chromatography (EtOAc in PE 20 to 45%) to afford (2-

(methylthio)benzo[d]thiazol-6-yl)methanol (598 mg, 71%) as a beige solid. MS (ES+) $C_9H_9NOS_2$ requires: 211, found: 212 $[M+H]^+$.

Step 3: 6-(chloromethyl)-2-(methylthio)benzo[d]thiazole. MsCl (572 mg, 5 mmol) was added to a solution of (2-(methylthio)benzo[d]thiazol-6-yl)methanol (350 mg, 1.66 mmol) and DIPEA (642 mg, 5 mmol) in DCM (10 mL) at 0°C. The mixture was treated with 3 drops of DMF and stirred at rt for 16h. $NaHCO_3$ (sat. aq. 15 mL) was added, and the aqueous phase was extracted with DCM (3 x 20 mL). The combined organic layers were washed with brine, dried over $MgSO_4$ and concentrated under reduced pressure to provide 6-(chloromethyl)-2-(methylthio)benzo[d]thiazole (380 mg, quant.) as a beige solid. MS (ES+) $C_9H_8ClNS_2$ requires: 229, found: 230[M+H]+.

Step 4: 2-(methylthio)-6-((pyridin-3-yloxy)methyl)benzo[d]thiazole. A solution of pyridine 3-ol (123 mg, 1.29 mmol) in DMA (1 mL) was added to a mixture of t-BuOK (171 mg, 1.5 mmol) in DMA (2 mL). It was stirred at rt for 1 h and subsequently heated to 100°C for 1 h. A solution of 6-(chloromethyl)-2-(methylthio)benzo[d]thiazole (229 mg, 1 mmol) in DMA (1 mL) was added. The mixture was stirred for 2 h. The reaction mixture was cooled to rt, it was diluted with water and extracted with EtOAc (3 × 15 mL). The

combined organic phases were washed with brine, dried over $MgSO_4$ and concentrated under reduced pressure to provide 2-(methylthio)-6-((pyridin-3-yloxy)methyl)benzo[d]thiazole (288 mg, quant). The product was used in the next step withouth further purification. MS (ES+) $C_{14}H_{12}N_2OS_2$ requires: 288, found: 289[M+H]+.

Step 5: 2-(methylsulfinyl)-6-((pyridin-3-yloxy)methyl)benzo[d]thiazole.

To a solution of provide 2-(methylthio)-6-((pyridin-3-yloxy)methyl)benzo[d]thiazole (288 mg, 1 mmol, crude) in DCM (3 mL) was added *m*-CPBA (202 mg, 1 mmol, 85%). The reaction mixture was stirred at rt for 2 h. The reaction was quenched with Na_2CO_3 (sat. aq. 5 mL), extracted with DCM (3 x 15 mL) and the combined organic phases were dried over $MgSO_4$and concentrated under reduced pressure to provide 2-(methylsulfinyl)-6-((pyridin-3-yloxy)methyl)benzo[d]thiazole (304 mg crude, 100 %). MS (ES+) $C_{14}H_{12}N_2O_2S_2$ requires: 304, found: 305[M+H]+.

Step 6: N-cyclohexyl-6-((pyridin-3-yloxy)methyl)benzo[d]thiazol-2-amine.

A mixture of 2-(methylsulfinyl)-6-((pyridin-3-yloxy)methyl)benzo[d]thiazole (50 mg, 0.16 mmol) in cyclohexanamine (0.5 mL) was stirred at 100°C for 2 h. The residue was purified

by preparative HPLC (Mobile phase: A = 0.1% NH_4HCO_3/H_2O, B = MeCN; Gradient: B = 5 - 95%; 12 min; Column: C18) to provide the product (10 mg, 18%).

MS (ES+) $C_{19}H_{21}N_3OS$ requires: 339, found: 340[M+H]+; MS (ES+) $C_{19}H_{21}N_3OS$ requires: 339, found: 340[M+H]+; 1H NMR (500 MHz, d_4-MeOD) δ 8.32 (d, J = 2.9 Hz, 1H), 8.14 (dd, J = 4.7, 0.6 Hz, 1H), 7.70 (d, J = 1.0 Hz, 1H), 7.52 (ddd, J = 8.5, 2.7, 0.8 Hz, 1H), 7.43 (d, J = 8.3 Hz, 1H), 7.40 – 7.34 (m, 2H), 5.20 (s, 2H), 3.78 – 3.71 (m, 1H), 2.14 – 2.07 (m, 2H), 1.86 – 1.80 (m, 2H), 1.73 – 1.67 (m, 1H), 1.52 – 1.42 (m, 2H), 1.39 – 1.26 (m, 3H).

Synthesis of N-cyclohexyl-6-((pyridin-4-yloxy)methyl)benzo[d]thiazol-2-amine (5)

N-cyclohexyl-6-((pyridin-4-yloxy)methyl)benzo[d]thiazol-2-amine (5) was prepared as described for N-cyclohexyl-6-((pyridin-3-yloxy)methyl)benzo[d]thiazol-2-amine (4).

MS (ES+) $C_{19}H_{21}N_3OS$ requires: 339, found: 340[M+H]+; 1H NMR (500 MHz, DMSO-d_6) δ 8.02 (d, J = 7.5 Hz, 1H), 7.79 – 7.71 (m, 2H), 7.65 (d, J = 1.6 Hz, 1H), 7.36 (d, J = 8.2 Hz, 1H), 7.19 (dd, J = 8.3, 1.8 Hz, 1H), 6.11 – 6.04 (m, 2H), 5.02 (s, 2H), 3.71 – 3.66 (m, 1H), 1.99 – 1.91 (m, 2H), 1.75 – 1.67 (m, 2H), 1.61 – 1.53 (m, 1H), 1.36 – 1.15 (m, 5H).

Synthesis of N-cyclohexyl-6-((pyridin-3-ylamino)methyl)benzo[d]thiazol-2-amine (6)

Step 1: 6-(azidomethyl)-2-(methylthio)benzo[d]thiazole. To a solution of (2-(methylthio)benzo[d]thiazol-6-yl)methanol (100 mg, 0.473 mmol) in THF (1578 µl) at 0 °C were added diphenyl phosphorazidate (122 µl, 0.568 mmol) and DBU (86 µl, 0.57 mmol) and the resulting mixture was stirred at rt for 3 h. The volatiles were removed under reduced pressure and the residue was purified via silica gel chromatography (0 to 100 % EtOAc in hexanes) to provide 6-(azidomethyl)-2-(methylthio)benzo[d]thiazole (103 mg, 92 % yield) as a colorless liquid that solidified overnight. MS (ES+) $C_9H_8N_4S_2$ requires: 236, found: NA.

Step 2: (2-(methylthio)benzo[d]thiazol-6-yl)methanamine. A reaction vessel was charged with 6-(azidomethyl)-2-(methylthio)benzo[d]thiazole (73 mg, 0.309 mmol), Lindlar's catalyst (66 mg, 0.031 mmol) and EtOH (3 mL) under an atmosphere of N_2. The suspension was degassed with N_2 for 5 min. and purged with H_2 for 5 min. The reaction mixture was stirred under an atmosphere of H_2 at 1 atm for 2 h. The reaction mixture was purged with N_2, filtered through Celite and concentrated under reduced pressure. The residue was purified via silica gel chromatography (0 - 10 % MeOH in DCM w/ 0.1%

NH_4OH) to provide (2-(methylthio)benzo[d]thiazol-6-yl)methanamine (70 mg, 81 % yield) as a yellow liquid. MS (ES+) $C_9H_{10}N_2S_2$ requires: 210, found: 211 [M+H] +.

Step 3: N-((2-(methylthio)benzo[d]thiazol-6-yl)methyl)pyridin-3-amine. To a suspension of 3-iodopyridine (49 mg, 0.24 mmol) and (2-(methylthio)benzo[d]thiazol-6-yl)methanamine (50 mg, 0.24 mmol) in DMSO-d_6 (475 µl) were added Cu (1.5 mg, 0.024 mmol) and CsOAc (91 mg, 0.47 mmol) and the resulting mixture was stirred at 90 °C for 7 h. The reaction was diluted with MeOH (1 mL), and TFA was added. The mixture was filtered, the solvent was removed under reduced pressure and the residue was purified by mass-triggered preparative HPLC (Mobile phase: A = 0.1% TFA/H_2O, B = 0.1% TFA/MeCN; Gradient: B = 10 - 90%; 12 min; Column: C18) to provide N-((2-(methylthio)benzo[d]thiazol-6-yl)methyl)pyridin-3-amine (53 mg, 78% yield) as a brown amorphous material. MS (ES+) $C_{14}H_{13}N_3S_2$ requires: 287, found: 288 [M+H] +.

Step 4: N-((2-(methylsulfinyl)benzo[d]thiazol-6-yl)methyl)pyridin-3-amine. To a suspension of N-((2-(methylthio)benzo[d]thiazol-6-yl)methyl)pyridin-3-amine (50 mg, 0.17 mmol) in DCM (1.7 mL) at 0 °C was added *m*CPBA (43 mg, 0.17 mmol) in DCM (0.5 mL) in portions and the resulting mixture was stirred at 0 °C for 1 h. The solution was

diluted with $NaHCO_3$ (aq., sat., 2 mL) and extracted with DCM (3 x 1 mL). The volatiles were removed under reduced pressure to provide N-((2-(methylsulfinyl)benzo[d]thiazol-6-yl)methyl)pyridin-3-amine (53 mg, 100 % yield) as a brown amorphous material. MS (ES+) $C_{14}H_{13}N_3OS_2$ requires: 303, found: 304 [M+H] +.

Step 5: N-cyclohexyl-6-((pyridin-3-ylamino)methyl)benzo[d]thiazol-2-amine. A microwave vial was charged with a solution of N-((2-(methylsulfinyl)benzo[d]thiazol-6-yl)methyl)pyridin-3-amine (21 mg, 0.069 mmol) in DMA (277 µl) and cyclohexanamine (23 µl, 0.21 mmol) and Hunig'sBase (12 µl, 0.069 mmol) was added. The resulting mixture was stirred at 120 °C for 24 h. The mixture was diluted with MeOH, acidified with TFA, and the residue was purified by mass-triggered preparative HPLC (Mobile phase: A = 0.1% TFA/H_2O, B = 0.1% TFA/MeCN; Gradient: B = 10 - 90%; 12 min; Column: C18) to provide N-cyclohexyl-6-((pyridin-3-ylamino)methyl)benzo[d]thiazol-2-amine (2 mg, 8.54 % yield) as a pale yellow amorphous material. MS (ES+) $C_{19}H_{22}N_4S$ requires: 338, found: 339 [M+H] +. ^{1}H NMR (500 MHz, DMSO-d_6) δ 8.25 (brs, 1H), 8.06 (d, J = 5.0 Hz, 1H), 8.03 (d, J = 5.0 Hz, 1H), 7.92 (s, 1H), 7.70 (m, 3H), 7.37 (d, J = 10.0 Hz, 1H), 7.24 (d, J = 5.0 Hz, 1H), 4.4 (s, 2H), 3.7(m, 1H), 1.71-1.76 (m, 4H), 1.25-1.4 (m, 6H).

Synthsis of N-cyclohexyl-6-((pyridin-4-ylamino)methyl)benzo[d]thiazol-2-amine (7)

N-cyclohexyl-6-((pyridin-4-ylamino)methyl)benzo[d]thiazol-2-amine was prepared as described for N-cyclohexyl-6-((pyridin-3-yloxy)methyl)benzo[d]thiazol-2-amine (6).

MS (ES+) $C_{19}H_{23}N_5O_3S$, requires:401, found: 402[M+H]+; 1H NMR (400 MHz, d_4-MeOD) δ 8.27 (d, J = 5.2 Hz, 1H), 6.86 (d, J = 5.1 Hz, 1H), 6.83 (d, J = 2.3 Hz, 1H), 6.63 (d, J = 2.2 Hz, 1H), 4.97 (s, 2H), 3.94 (s, 3H), 3.66 – 3.54 (m, 1H), 3.46 – 3.37 (m, 1H), 2.20 – 2.10 (m, 1H), 2.07 – 2.00 (m, 1H), 1.80 – 1.71 (m, 2H), 1.44 – 1.31 (m, 4H).

Synthesis of (1R,2R)-2-(6-((2-Aminopyrimidin-4-yl)methoxy)-4-methoxybenzo[d]thiazol-2-ylamino)cyclohexanol (21)

Step 1: 2-Chloro-4-(chloromethyl)pyrimidine. To a solution of 2-chloro-4-methylpyrimidine (5.0 g, 39 mmol) in CCl_4 (100 ml) was added NCS (7.8 g, 58 mmol) and AIBN (0.64 g, 3.9 mmol) and the resulting mixture was heated at reflux for 18 h. The reaction mixture was allowed to cool to rt. The reaction mixture was filtered through a Celite pad, and the filtrate was concentrated under reduced pressure. The residue was purified via silica gel chromatography (10 to 40% EtOAc in hexanes) to give 2-chloro-4-

(chloromethyl)pyrimidine (3.1 g, 49%) as a clear oil. MS (ES+) $C_5H_4Cl_2N_2$ requires: 162, found: 163 $[M+H]^+$.

Step 2: (1R,2R)-2-(6-((2-Chloropyrimidin-4-yl)methoxy)-4-methoxybenzo[d]thiazol-2-ylamino)cyclohexanol. A mixture of 2-((1R,2R)-2-hydroxycyclohexylamino)-4-methoxybenzo[d]thiazol-6-ol (40 mg, 0.14 mmol) and Cs_2CO_3 (116 mg, 0.36 mmol) in DMF (2 mL) was stirred at rt for 1 h. 2-Chloro-4-(chloromethyl)pyrimidine (30 mg, 0.19 mmol) was added and the mixture was stirred at rt for 4 h. The mixture was diluted with water, extracted with EtOAc (3 x 30 mL), the combined organic phases were washed with brine, dried over Na_2SO_4 and concentrated under reduced pressure to provide the product (38 mg, 67%) as a brown solid. MS (ES+) $C_{19}H_{21}ClN_4O_3S$, requires: 421, found: 421, 423 $[M+H]^+$.

Step 3: (1R,2R)-2-(6-((2-Aminopyrimidin-4-yl)methoxy)-4-methoxybenzo[d]thiazol-2-ylamino)cyclohexanol. A mixture of (1R,2R)-2-(6-((2-chloropyrimidin-4-yl)methoxy)-4-methoxybenzo[d]thiazol-2-ylamino)cyclohexanol (38 mg, 0.095 mmol) in NH_4OH (aq., sat.) solution was stirred at 90°C for 3 h in a sealed tube. The mixture was cooled to rt, concentrated and purified by prep-HPLC (NH_4HCO_3) to provide the product (3 mg, 8%)

as a beige solid. MS (ES+) $C_{19}H_{23}N_5O_3S$, requires: 401, found: 402 [M+H]+; ^{1}H NMR (400 MHz, d_4-MeOD) δ 8.27 (d, J = 5.2 Hz, 1H), 6.86 (d, J = 5.1 Hz, 1H), 6.83 (d, J = 2.3 Hz, 1H), 6.63 (d, J = 2.2 Hz, 1H), 4.97 (s, 2H), 3.94 (s, 3H), 3.66 – 3.54 (m, 1H), 3.46 – 3.37 (m, 1H), 2.20 – 2.10 (m, 1H), 2.07 – 2.00 (m, 1H), 1.80 – 1.71 (m, 2H), 1.44 – 1.31 (m, 4H).

Synthesis of (1S,2S)-2-((6-((2-Aminopyrimidin-4-yl)methoxy)-4-methoxybenzo[d]thiazol-2-yl)amino)cyclohexan-1-ol (22)

Step 1: 2-Chloro-4-((3-methoxy-4-nitrophenoxy)methyl)pyrimidine. To a solution of 3-methoxy-4-nitrophenol (2.83 g, 16.7 mmol) and 2-chloro-4-(chloromethyl)pyrimidine (3.0 g, 18 mmol) in DMF (16 ml) was added K_2CO_3 (2.8 g, 20 mmol) and the resulting mixture was stirred at rt for 20 h. Ice cold water (30 ml) was added and the mixture was stirred for 10 min before filtering on a Buchner funnel. The solid was washed with cold water (10 mL) and dried under vacuum for 2 h to give 2-chloro-4-((3-methoxy-4-nitrophenoxy)methyl)pyrimidine (4.6 g, 93%) as a light brown solid. MS (ES+) $C_{12}H_{10}ClN_3O_4$ requires: 295, found: 296 $[M+H]^+$.

Step 2: 4-((2-Chloropyrimidin-4-yl)methoxy)-2-methoxyaniline. A reaction vessel was charged with 2-chloro-4-((3-methoxy-4-nitrophenoxy)methyl)pyrimidine (4.6 g, 16 mmol), 10% Pt-C (460 mg, 0.12 mmol), and THF-MeOH (5:1, 60 ml). The suspension was degassed with N_2 for 3 min and purged with H_2 for 3 min. The reaction mixture was stirred under an atmosphere of H_2 at 1 atm for 2 h. The reaction mixture was purged with N_2, filtered through Celite, and concentrated under reduced pressure to give 4-((2-chloropyrimidin-4-yl)methoxy)-2-methoxyaniline (4.0 g, 97%) as a light yellow powder. MS (ES+) $C_{12}H_{12}ClN_3O_2$ requires: 265, found: 266 $[M+H]^+$.

Step 3: 1-(4-((2-Chloropyrimidin-4-yl)methoxy)-2-methoxyphenyl)-3-((1S,2S)-2-hydroxycyclohexyl)thiourea. To a solution of 4-((2-chloropyrimidin-4-yl)methoxy)-2-methoxyaniline (4.0 g, 15 mmol) in DCM (200 ml) was added di(1H-imidazol-1-yl)methanethione (3.2 g, 18 mmol) and the resulting mixture was stirred at rt for 4 h. (1S,2S)-2-Aminocyclohexanol (3.5 g, 30 mmol) was added and the resulting mixture was stirred at rt for 3 h. The volatiles were removed under reduced pressure. The residue was purified via silica gel chromatography (25 - 100% EtOAc in hexanes) to give 1-(4-((2-chloropyrimidin-4-yl)methoxy)-2-methoxyphenyl)-3-((1S,2S)-2-

hydroxycyclohexyl)thiourea (6.1 g, 96%) as a white solid. MS (ES+) $C_{19}H_{23}ClN_4O_3S$ requires: 422, found: 423 $[M+H]^+$.

Step 4: (1S,2S)-2-((6-((2-Chloropyrimidin-4-yl)methoxy)-4-methoxybenzo[d]thiazol-2-yl)amino)cyclohexan-1-ol. To a solution of 1-(4-((2-chloropyrimidin-4-yl)methoxy)-2-methoxyphenyl)-3-((1S,2S)-2-hydroxycyclohexyl)thiourea (1.0 g, 2.3 mmol) in DCM (100 ml) was added BSTFA (1.3 ml, 4.7 mmol) and the resulting solution was stirred for 5 min. Solid benzyltrimethylammonium tribromide (0.92 g, 2.3 mmol) was added into the previous reaction mixture. After 20 min sat. $NaHCO_3$ (30 ml) was added and the reaction mixture was stirred for 5 min. The biphasic mixture was transferred to a separatory funnel with an additional 20 ml of DCM, and the layers were separated. The organic layers were washed with saturated aq. $NaHCO_3$ (30 ml), followed by 10% aq. $Na_2S_2O_3$ (1 x 20 ml), dried over $MgSO_4$, filtered, and concentrated under reduced pressure. The residue was triturated with 1:2 Ether/Hexanes (30 ml) and the resulting solid was filtered through a Buchner funnel. After rinsing with 1:1 Ether/hexanes (10 ml), (1S,2S)-2-((6-((2-chloropyrimidin-4-yl)methoxy)-4-methoxybenzo[d]thiazol-2-yl)amino)cyclohexan-1-ol

was collected as off white powder (945 mg, 95%). MS (ES+) $C_{19}H_{21}ClN_4O_3S$ requires: 420, found: 421 $[M+H]^+$.

Step 5: (1S,2S)-2-((6-((2-Aminopyrimidin-4-yl)methoxy)-4-methoxybenzo[d]thiazol-2-yl)amino)cyclohexan-1-ol. A microwave vial was charged with (1S,2S)-2-((6-((2-chloropyrimidin-4-yl)methoxy)-4-methoxybenzo[d]thiazol-2-yl)amino)cyclohexanol (300 mg, 0.71 mmol) and NH_3 (7M in MeOH, 2.0 mL). The vial was sealed and the reaction mixture was heated to 120 °C in the microwave reactor for 4 h. The volatiles were removed under reduced pressure. The residue was purified by reverse phase preparative HPLC (Mobile phase: A = 0.1% TFA/H_2O, B = 0.1% TFA/MeCN; Gradient: B = 10 - 90%; 20 min; Column: C18) to give (1S,2S)-2-((6-((2-aminopyrimidin-4-yl)methoxy)-4-methoxybenzo[d]thiazol-2-yl)amino)cyclohexan-1-ol as a TFA salt (182 mg, 64%) as an off white powder. MS (ES+) $C_{19}H_{23}N_5O_3S$ requires: 401, found: 402 $[M+H]^+$; ^1H NMR (600 MHz, DMSO-d_6) δ 8.27 (d, J = 6.5 Hz, 1H), 6.86 (d, J = 5.2 Hz, 1H), 6.83 (d, J = 2.4 Hz, 1H), 6.62 (d, J = 2.0 Hz, 1H), 4.90 (s, 2H), 3.93 (s, 3H), 3.59 (m, 1H), 3.40 (m, 1H), 2.14 (m, 1H), 2.02 (m, 1H), 1.76 (m, 2H), 1.43-1.28 (m, 4H).

Synthesis of (1S,2S)-2-((6-((2-Aminopyrimidin-4-yl)methoxy)-7-chloro-4-methoxybenzo[d]thiazol-2-yl)amino)cyclohexan-1-ol (23)

Step 1: 2-Chloro-5-methoxy-4-nitrophenol. To a solution of 3-methoxy-4-nitrophenol (1.0 g, 5.9 mmol) at 0 °C was added NCS (0.868g, 6.50 mmol), and the resulting mixture was stirred at 50°C for 2 h. The reaction mixture was diluted with EtOAc (20 mL) and washed with water (2 x 20 mL). The layers were separated, and the organic layer was washed with sat. NaCl (10 mL), dried over $MgSO_4$, filtered and concentrated under reduced pressure to provide 2-chloro-5-methoxy-4-nitrophenol (1.2 g, ~100%) as a yellow solid. MS (ES+) $C_7H_6ClNO_4$ requires: 203, found: 204 $[M+H]^+$.

Step 2-6: As described for compound 22. MS (ES+) $C_{19}H_{22}ClN_5O_3S$ requires: 435, found: 436 $[M+H]^+$; ^1H NMR (600 MHz, DMSO-d_6) δ 8.27 (d, J = 5.0 Hz, 1H), 7.89 (d, J = 7.7 Hz, 1H), 6.79 (s, 1H), 6.74 (d, J = 4.9 Hz, 1H), 6.67 (s, 2H), 5.04 (s, 2H), 4.75 (d, J = 5.1 Hz, 1H), 3.85 (s, 3H), 3.53 – 3.46 (m, 1H), 3.36 – 3.32 (m, 1H), 2.09 – 1.99 (m, 1H), 1.92 – 1.84 (m, 1H), 1.69 – 1.56 (m, 2H), 1.31 – 1.16 (m, 4H).

Synthesis of (1R,2R)-2-(6-((2-aminopyridin-4-yl)methoxy)-4-methoxybenzo[d]thiazol-2-ylamino)cyclohexanol (24)

Step 1: Tert-butyl 4-((2-((1R,2R)-2-hydroxycyclohexylamino)-4-methoxybenzo[d]thiazol-6-yloxy)methyl)pyridin-2-ylcarbamate. A solution of 2-((1R,2R)-2-hydroxycyclohexylamino)-4-methoxybenzo[d]thiazol-6-ol (100 mg, 0.34 mmol) tert-butyl 4-(chloromethyl)pyridin-2-ylcarbamate (83 mg, 0.34 mmol) and Cs_2CO_3 (333 mg, 1.02 mmol) in DMF (2 mL) was stirred at 80°C for 2 h. The reaction mixture was diluted with water (5 mL) and extracted with EtOAc (3 x 50 mL). The combined organic phases were washed with water (10 ml), brine (10 ml), dried over Na_2SO_4, filtered and concentrated under reduced pressure to provide tert-butyl 4-((2-((1R,2R)-2-hydroxycyclohexylamino)-4-methoxybenzo[d]thiazol-6-yloxy)methyl)pyridin-2-ylcarbamate (30 mg, 17%) as a yellow solid. MS (ES+) $C_{25}H_{32}N_4O_5S$ requires: 500, found: 501[M+H]+.

Step 2: (1R,2R)-2-(6-((2-Aminopyridin-4-yl)methoxy)-4-methoxybenzo[d]thiazol-2-ylamino)cyclohexanol. A mixture of tert-butyl 4-((2-((1R,2R)-2-hydroxycyclohexylamino)-4-methoxybenzo[d]thiazol-6-yloxy)methyl)pyridin-2-ylcarbamate (30 mg, 0.06 mmol) in TFA (1mL) was stirred at rt for 16 h. The volatiles were removed under reduced pressure and the residue was purified by prep-HPLC to provide (1R,2R)-2-(6-((2-aminopyridin-4-

yl)methoxy)-4-methoxybenzo[d]thiazol-2-ylamino)cyclohexanol (8.6 mg, 35%) as a white solid. MS (ES+) $C_{21}H_{23}N_5O_2$ requires: 400, found: 401$[M+H]^+$; ^{1}H NMR (500 MHz, DMSO-d_6) δ 7.87 (d, J = 5.2 Hz, 1H), 7.62 (d, J = 7.5 Hz, 1H), 6.90 (d, J = 2.3 Hz, 1H), 6.54 – 6.48 (m, 3H), 5.97 (s, 2H), 4.95 (s, 2H), 4.78 (d, J = 5.0 Hz, 1H), 3.82 (s, 3H), 3.55 – 3.47 (m, 1H), 2.07 – 2.01 (m, 1H), 1.91 – 1.84 (m, 1H), 1.63 (t, J = 13.1 Hz, 2H), 1.32 – 1.16 (m, 4H).

Synthesis of N-(4-(((2-(((1R,2R)-2-hydroxycyclohexyl)amino)-4-methoxybenzo[d]thiazol-6-yl)oxy)methyl)pyridin-2-yl)acetamide (25)

Step 1: 2-bromo-4-methoxybenzo[d]thiazol-6-ol. A vial was charged with 6-(benzyloxy)-2-bromo-4-methoxybenzo[d]thiazole (0.80g, 2.28 mmol) and TFA (1.76 ml, 22.8 mmol) and the suspension stirred at 65°C for 40h. The volatiles were removed under reduced pressure. The residue was purified via silica gel chromatography (0 to 100 % EtOAc in hexanes), however, the product was contaminated with starting material. The solid was triturated with DCM (2 x 5 mL), centrifuged, and the liquid decanted. The remaining solid was dried to provide 2-bromo-4-methoxybenzo[d]thiazol-6-ol (142 mg, 0.546 mmol, 24% yield) as a white solid. MS (ES+) $C_8H_6BrNO_2S$ requires: 259 found: 259.9 $[M+H]^+$.

Step 2: Tert-butyl (4-(((2-bromo-4-methoxybenzo[d]thiazol-6-yl)oxy)methyl)pyridin-2-yl)carbamate. To a suspension of tert-butyl (4-(hydroxymethyl)pyridin-2-yl)carbamate (133 mg, 0.592 mmol), 2-bromo-4-methoxybenzo[d]thiazol-6-ol (140 mg, 0.538 mmol), and Ph_3P (198 mg, 0.754 mmol) in THF (2.6 mL) at 0 °C was added (E)-di-tert-butyl diazene-1,2-dicarboxylate (174 mg, 0.754 mmol) and the resulting mixture was stirred at rt for 16 h. The volatiles were removed under reduced pressure. The residue was purified via silica gel chromatography (0 to 100 % EtOAc in hexanes) to provide tert-butyl (4-(((2-bromo-4-methoxybenzo[d]thiazol-6-yl)oxy)methyl)pyridin-2-yl)carbamate (233 mg, 0.350 mmol, 93 % yield) as a pale yellow solid.

MS (ES+) C19H20BrN3O4S requires: 465 found: 466 $[M+H]^+$.

Step 3: N-(4-(((2-bromo-4-methoxybenzo[d]thiazol-6-yl)oxy)methyl)pyridin-2-yl)acetamide.

To a suspension of tert-butyl (4-(((2-bromo-4-methoxybenzo[d]thiazol-6-yl)oxy)methyl)pyridin-2-yl)carbamate (233 mg, 0.50 mmol) in DCM (1 mL) at 0 °C was added TFA (962 µl, 12.5 mmol) and the resulting mixture was stirred at rt for 3 h. The residue was adsorbed onto Celite and purified via flash chromatography (0 - 10 % MeOH

in DCM w/ 0.5% NH_4OH) to provide 4-(((2-bromo-4-methoxybenzo[d]thiazol-6-yl)oxy)methyl)pyridin-2-amine (109 mg, 0.298 mmol, 60%) as a green amorphous material. MS (ES+) $C_{14}H_{12}BrN_3O_2S$ requires: 365 found: 366 $[M+H]^+$.

Step 4: N-(4-(((2-bromo-4-methoxybenzo[d]thiazol-6-yl)oxy)methyl)pyridin-2-yl)acetamide. To a suspension of 4-(((2-bromo-4-methoxybenzo[d]thiazol-6-yl)oxy)methyl)pyridin-2-amine (69 mg, 0.19 mmol) and pyridine (76 µl, 0.942 mmol) in DMF (377 µl) was added Ac_2O (89 µl, 0.94 mmol) and the resulting mixture was stirred at 60°C for 1 h. The volatiles were removed under reduced pressure. The residue was purified via silica gel chromatography (0 - 10 % MeOH in DCM w/ 0.5% NH_4OH) to provide N-(4-(((2-bromo-4-methoxybenzo[d]thiazol-6-yl)oxy)methyl)pyridin-2-yl)acetamide (68 mg, 88%) as a green foam solid.

MS (ES+) $C_{16}H_{14}BrN_3O_3S$ requires: 407 found: 408 $[M+H]^+$.

Step 5: N-(4-(((2-(((1R,2R)-2-hydroxycyclohexyl)amino)-4-methoxybenzo[d]thiazol-6-yl)oxy)methyl)pyridin-2-yl)acetamide. To a solution of N-(4-(((2-bromo-4-methoxybenzo[d]thiazol-6-yl)oxy)methyl)pyridin-2-yl)acetamide (68 mg, 0.17 mmol) and (1R,2R)-2-aminocyclohexanol (57 mg, 0.50 mmol) in DMA (555 µl) was added DIPEA (32

µl, 0.183 mmol) and the resulting mixture was stirred at 100 °C for 12h. The volatiles were removed under reduced pressure. The residue was purified via silica gel chromatography (0 - 10 % MeOH in DCM w/ 0.5% NH_4OH) to provide N-(4-(((2-(((1R,2R)-2-hydroxycyclohexyl)amino)-4-methoxybenzo[d]thiazol-6-yl)oxy)methyl)pyridin-2-yl)acetamide (28 mg, 0.063 mmol, 38.0%) as a off-white solid. MS (ES+) $C_{22}H_{26}N_4O4_S$ requires: 442 found: 443 $[M+H]^+$. ^1H NMR (600 MHz, DMSO-d_6) δ 8.28 (d, J = 6.0 Hz, 1H), 7.63 (s, 1H), 7.14 (d, J = 6.0 Hz, 1H), 6.94 (d, J = 6.0 Hz, 1H), 6.56 (s, 1H), 5.13 (s, 2H), 4.78 (s, 1H), 3.83 (s, 3H), 3.5 (m, 1H), 2.09 (s, 3H), 2.03 (m, 1H), 1.86 (m, 1H), 1.63 (m, 2H), 1.23 (m, 4H).

Synthesis of 1-(4-(((2-(((1S,2S)-2-hydroxycyclohexyl)amino)-4-methoxybenzo[d]thiazol-6-yl)oxy)methyl)pyrimidin-2-yl)-3-methylurea (26)

Step 1: N-((1S,2S)-2-((tert-butyldimethylsilyl)oxy)cyclohexyl)-6-((2-chloropyrimidin-4-yl)methoxy)-4-methoxybenzo[d]thiazol-2-amine. To a solution of (1S,2S)-2-((6-((2-chloropyrimidin-4-yl)methoxy)-4-methoxybenzo[d]thiazol-2-yl)amino)cyclohexanol (300 mg, 0.72 mmol) in DMF (2 ml) were added TBDMSCl (130 mg, 0.86 mmol) and imidazole (150 mg, 2.2 mmol) and the resulting mixture was stirred at rt for 12 h. The residue was

purified via silica gel chromatography (10 to 50% EtOAc in hexanes to provide N-((1S,2S)-2-((tert-butyldimethylsilyl)oxy)cyclohexyl)-6-((2-chloropyrimidin-4-yl)methoxy)-4-methoxybenzo[d]thiazol-2-amine (347 mg, 91 % yield) as an off-white solid. MS (ES+) $C_{25}H_{35}ClN_4O_3SSi$ requires: 535, found: 536 [M+H] +.

Step 2: 6-((2-aminopyrimidin-4-yl)methoxy)-N-((1S,2S)-2-((tert-butyldimethylsilyl)oxy)cyclohexyl)-4-methoxybenzo[d]thiazol-2-amine. A microwave vial was charged with N-((1S,2S)-2-((tert-butyldimethylsilyl)oxy)cyclohexyl)-6-((2-chloropyrimidin-4-yl)methoxy)-4-methoxybenzo[d]thiazol-2-amine (300 mg, 0.56 mmol), and NH_4OH (aq., 2M) in 2-propanol (3 ml) was added. The vial was sealed and the reaction mixture was heated to 120 °C in the microwave reactor for 8 h. The volatiles were removed under reduced pressure. The residue was purified via silica gel chromatography (20 to 70% EtOAc in hexanes to provide 6-((2-aminopyrimidin-4-yl)methoxy)-N-((1S,2S)-2-((tert-butyldimethylsilyl)oxy)cyclohexyl)-4-methoxybenzo[d]thiazol-2-amine (208 mg, 72%) as an off white solid. MS (ES+) $C_{25}H_{37}N_5O_3SSi$ requires: 515, found: 516 [M+H] +.

Step 3: 1-(4-(((2-(((1S,2S)-2-((tert-butyldimethylsilyl)oxy)cyclohexyl)amino)-4-methoxybenzo[d]thiazol-6-yl)oxy)methyl)pyrimidin-2-yl)-3-methylurea. To a solution of 6-((2-aminopyrimidin-4-yl)methoxy)-N-((1S,2S)-2-((tert-butyldimethylsilyl)oxy)cyclohexyl)-4-methoxybenzo[d]thiazol-2-amine (100 mg, 0.20 mmol) in DCM (5 ml) were added pyridine (33 uL, 0.39 mmol), DMAP (2 mg, 0.02 mmol) and phenyl chloroformate (40 mg, 0.25 mmol). The resulting mixture was stirred at rt for 3 h. The reaction was quenched with $MeNH_2$ (0.2 ml, 1.4 mmol) (7N in MeOH) and stirred for 16 h. The volatiles were removed under reduced pressure and the residue was diluted with EtOAc (15 mL), $NaHCO_3$ (aq. 10w/w%, 5 mL) was added, and the layers were separated. The aqueous phase was extracted with EtOAc (1 x 5 mL), the combined organic layers were washed with $NaHCO_3$ (aq. 10w/w%, 5 mL), dried over $MgSO_4$, filtered and concentrated under reduced pressure. The residue was purified via silica gel chromatography (20 to 80 % EtOAc in hexane to provide 1-(4-(((2-(((1S,2S)-2-((tert-butyldimethylsilyl)oxy)cyclohexyl)amino)-4-methoxybenzo[d]thiazol-6-yl)oxy)methyl)pyrimidin-2-yl)-3-methylurea (57 mg, 51%) as a yellow oil. MS (ES+) $C_{27}H_{40}N_6O_4SSi$ requires: 572, found: 573 [M+H] +.

Step 4: 1-(4-(((2-(((1S,2S)-2-hydroxycyclohexyl)amino)-4-methoxybenzo[d]thiazol-6-yl)oxy)methyl)pyrimidin-2-yl)-3-methylurea. To a solution of 1-(4-(((2-(((1S,2S)-2-((tert-butyldimethylsilyl)oxy)cyclohexyl)amino)-4-methoxybenzo[d]thiazol-6-yl)oxy)methyl)pyrimidin-2-yl)-3-methylurea (50 mg, 0.087 mmol) in DCM (2ml) was added TBAF (0.3 mL, 0.3 mmol) and the resulting mixture was stirred at rt for 12 h. The volatiles were removed under reduced pressure. The residue was purified by mass-triggered preparative HPLC (Mobile phase: A = 0.1% TFA/H2O, B = 0.1% TFA/MeCN; Gradient: B = 10- 50%; 20 min; Column: C18) to provide 1-(4-(((2-(((1S,2S)-2-hydroxycyclohexyl)amino)-4-methoxybenzo[d]thiazol-6-yl)oxy)methyl)pyrimidin-2-yl)-3-methylurea (16 mg, 40%) as a white solid. MS (ES+) $C_{21}H_{26}N_6O_4S$ requires: 458.5, found: 459.5 [M+H] +. ^{1}H NMR (600 MHz, DMSO-d_6) δ 10.49 (s, 1H), 8.64 (d, J = 5.4 Hz, 1H), 7.24 (d, J = 5.4 Hz, 1H), 7.18 (s, 1H), 7.1 (s, 1H),7.02 (m, 1H), 6.68 (s, 1H), 5.09 (s, 2H), 3.87 (s, 3H), 3.66 (s, 3H), 3.56 (m, 1H), 3.31-3.35 (m, 1H), 2.01 (m, 1H), 1.88 (m, 1H), 1.64 (m, 2H), 1.2-1.32 (m, 4H).

Synthesis of (1S,2S)-2-(7-chloro-4-methoxy-6-((2-(1-methyl-1H-pyrazol-4-ylamino)pyrimidin-4-yl)methoxy)benzo[d]thiazol-2-ylamino)cyclohexanol IACS-9439 (1)

Step 1: – (1S,2S)-2-((4-methoxy-6-((2-((1-methyl-1H-pyrazol-4-yl)amino)pyrimidin-4-yl)methoxy)benzo[d]thiazol-2-yl)amino)cyclohexan-1-ol. A microwave vial was charged with (1S,2S)-2-((6-((2-chloropyrimidin-4-yl)methoxy)-4-methoxybenzo[d]thiazol-2-yl)amino)cyclohexanol (550 mg, 1.31 mmol), 1-methyl-1H-pyrazol-4-amine (381 mg, 3.92 mmol) and DIPEA (456 µl, 2.61 mmol) and 2-propanol (5.2 mL) was added. The vial was sealed and the reaction mixture was heated to 120 °C in the microwave reactor for 5 h. The volatiles were removed under reduced pressure. The residue was purified by reverse phase preparative HPLC (Mobile phase: A = 0.1% NH_4OH/H_2O, B = 0.1% NH_4OH/MeCN; Gradient: B = 10 - 90%; 20 min; Column: C18) to provide the title compound (290 mg, 46%) as a light tan foam solid. HRMS (ES+) $C_{23}H_{28}N_7O_3S^+$ requires: 482.1969, found: 482.1971 $[M+H]^+$. ^{1}H NMR (500 MHz, DMSO-d_6) δ 9.50 (s, 1H), 8.41 (d, J = 3.6 Hz, 1H), 7.88 (s, 1H), 7.61 (d, J = 6.0 Hz, 2H), 7.47 (s, 1 H), 6.95 (s, 1H), 6.82 - 6.89 (d, J = 4.0, 1H), 6.59 (s, 1H), 5.0.5 (s, 2H), 4.76 (d, J = 4.0 Hz, 1H), 3.83 (s, 3H), 3.78 (s, 3H), 3.5 (m, 1H), 2.05 (m, 1H), 1.86 (m, 1H), 1.62 (m, 2H), 1.29 - 1.16 (m, 4H). ^{13}C NMR (151 MHz, DMSO-d_6) δ 166.6,0 163.71, 159.18, 158.69, 153.44, 149.98, 136.38, 131.27,

129.77, 123.08, 120.45, 107,53, 98.11, 97.73, 71.49, 69.62, 59.26, 55.62, 39.92, 39.78, 39.64, 39.51, 39.37, 39.23, 39.09, 38.61, 30.59, 23.91, 23.51.

Synthesis of (1S,2S)-2-((4-methoxy-6-((2-((1-methyl-1H-pyrazol-3-yl)amino)pyrimidin-4-yl)methoxy)benzo[d]thiazol-2-yl)amino)cyclohexan-1-ol (27)

Step 1: (1S,2S)-2-((4-methoxy-6-((2-((1-methyl-1H-pyrazol-3-yl)amino)pyrimidin-4-yl)methoxy)benzo[d]thiazol-2-yl)amino)cyclohexan-1-ol. A solution of (1S,2S)-2-((6-((2-chloropyrimidin-4-yl)methoxy)-4-methoxybenzo[d]thiazol-2-yl)amino)cyclohexanol (20 mg, 0.048 mmol), 1-methyl-1H-pyrazol-3-amine (46 mg, 0.47 mmol) and DIPEA (0.2 mL) in DMA (0.5 ml) was stirred at 110°C for 18 h. The residue was purified by mass-triggered preparative HPLC (Mobile phase: A = 0.1% TFA/H_2O, B = 0.1% TFA/MeCN; Gradient: B = 10 - 50%; 20 min; Column: C18) to provide (1S,2S)-2-((4-methoxy-6-((2-((1-methyl-1H-pyrazol-3-yl)amino)pyrimidin-4-yl)methoxy)benzo[d]thiazol-2-yl)amino)cyclohexanol (3.5 mg, 15% yield) as a brown solid. MS (ES+) $C_{23}H_{27}N_7O_3S$ requires: 481, found: 482.5 [M+H] +. 1H NMR (600 MHz, d_4-MeOD) δ 8.4 (s, 1H), 7.48 (s, 1H), 7.02 (d, J = 5.4 Hz, 1H), 6.99 (s, 1H), 6.84 (s, 1H), 6.53 (s, 1H), 5.14 (s, 2H), 3.99 (s, 3H), 3.84 (s, 3H), 3.65 (m, 1H), 3.42-3.49 (m, 1H), 2.05 – 2.1 (m, 2H), 1.78 (m, 2H), 1.33-1.42 (m, 4H).

Synthesis of (1S,2R)-2-((6-((2-((1H-pyrazol-4-yl)amino)pyrimidin-4-yl)methoxy)-4-methoxybenzo[d]thiazol-2-yl)amino)cyclohexanol (28)

Step 1: (1S,2R)-2-((6-((2-((1H-pyrazol-4-yl)amino)pyrimidin-4-yl)methoxy)-4-methoxybenzo[d]thiazol-2-yl)amino)cyclohexanol: A microwave vial was charged with (1S,2S)-2-((6-((2-chloropyrimidin-4-yl)methoxy)-4-methoxybenzo[d]thiazol-2-yl)amino)cyclohexanol (25 mg, 0.06 mmol), 1*H*-pyrazol-4-amine (20 mg, 0.24 mmol), TEA (0.025 mL, 0.18 mmol) and DMSO-d_6 (1.5 mL). The reaction mixture was heated to 120°C in the microwave reactor for 2 h. The mixture was purified by mass-triggered preparative HPLC (Mobile phase: A = 0.1% TFA/H2O, B = 0.1% TFA/MeCN; Gradient: B = 20 - 50%; 12 min; Column: C18) to give (1S,2R)-2-((6-((2-((1H-pyrazol-4-yl)amino)pyrimidin-4-yl)methoxy)-4-methoxybenzo[d]thiazol-2-yl)amino)cyclohexanol (12.2 mg, 0.026 mmol, 43.9 % yield) as a gray solid. MS (ES+) $C_{22}H_{25}N_7O_3S$ requires: 467.5, found: 468.5 [M+H]+. ^{1}H NMR (600 MHz, d_4-MeOD) δ 8.38 (brs, 1H), 8.15 (m, 2H), 7.0 (s, 1H), 6.92 (s, 1H), 6.86 (s, 1H), 5.13 (s, 2H), 3.99 (s, 3H), 3.60 (m, 1H), 3.47-3.50 (m, 1H), 2.05 – 2.09 (m, 2H), 1.79 (m, 2H), 1.32-1.48 (m, 4H).

(1S,2S)-2-(6-((2-(1H-pyrazol-5-ylamino)pyrimidin-4-yl)methoxy)-4-methoxybenzo [d]thiazol-2-ylamino)cyclohexanol (29)

Step 1: (1S,2S)-2-(6-((2-(1H-pyrazol-5-ylamino)pyrimidin-4-yl)methoxy)-4-methoxybenzo [d]thiazol-2-ylamino)cyclohexanol. A mixture of (1S,2S)-2-(6-((2-chloropyrimidin-4-yl)methoxy)-4-methoxybenzo[d]thiazol -2-ylamino)cyclohexanol (45 mg, 0.11 mmol), 1H-pyrazol-3-amine (11 mg, 0.13 mmol), $Pd_2(dba)_3$ (10 mg, 0.011 mmol), Xantphos (6 mg, 0.011) and Cs_2CO_3 (72 mg, 0.22 mmol) in dioxane (2 ml) was stirred at 110°C for 2 h under N_2 atmosphere. The solids were filtered and the filtrate was concentrated under reduced pressure. The residue was purified by preparative HPLC (NH_4HCO_3) to provide the title compound (6 mg, 14%) as a white solid. MS (ES+) $C_{22}H_{25}N_7O_3S$ requires: 467 found: 468 $[M+H]^+$. ^1H NMR (500 MHz, d_4-MeOD) δ 8.68 (d, J = 5 Hz, 1H), 8.41 (d, *J* = 3 Hz, 1H), 7.40 (d, *J* = 5 Hz, 1H), 6.90 (m, d, *J* = 2 Hz, 1H), 6.67 (d, *J* = 3 Hz, 1H), 6.00 (d, *J* = 2.5 Hz, 1H), 5.20 (s, 2H), 3.95 (s, 3H), 3.60-3.61 (m, 1H), 3.41-3.42 (m, 1H), 2.14-2.16 (m, 1H), 2.03-2.05 (m, 1H), 1.72-1.79 (m, 2H), 1.31-1.43 (m, 4H).

Synthesis of (1S,2S)-2-((6-((2-(isoxazol-4-ylamino)pyrimidin-4-yl)methoxy)-4-methoxybenzo[d]thiazol-2-yl)amino)cyclohexan-1-ol (30)

Step 1: (1S,2S)-2-((6-((2-(isoxazol-4-ylamino)pyrimidin-4-yl)methoxy)-4-methoxybenzo[d]thiazol-2-yl)amino)cyclohexan-1-ol. A vial was charged with (1S,2S)-2-((6-((2-chloropyrimidin-4-yl)methoxy)-4-methoxybenzo[d]thiazol-2-yl)amino)cyclohexanol (20 mg, 0.048 mmol), isoxazol-4-amine (18 mg, 0.14 mmol) (0.020 mL, 0.14 mmol) and MeOH (2mL) and the reaction was heated for 3 h at 100°C. The residue was purified by mass-triggered preparative HPLC (Mobile phase: A = 0.1% TFA/H_2O, B = 0.1% TFA/MeCN; Gradient: B = 20 - 50%; 12 min; Column: C18) to provide (1S,2S)-2-((6-((2-(isoxazol-4-ylamino)pyrimidin-4-yl)methoxy)-4-methoxybenzo[d]thiazol-2-yl)amino)cyclohexanol (14 mg, 0.030 mmol, 63%) as a light brown solid. MS (ES+) $C_{22}H_{24}N_6O_4S$ requires: 468.5, found: copy in m/z [M+H] +.

^{1}H NMR (600 MHz, d_4-MeOD) δ 9.06 (s, 1H), 8.52 (s, 1H), 8.46 (d, J = 5.4 Hz, 1H), 6.99 (dd, J = 15.0 and 5.4 Hz, 2H), 6.87 (s, 1H), 5.16 (s, 2H), 3.99 (s, 3H), 3.60 (m, 1H), 3.47-3.51 (m, 1H), 2.05 – 2.1 (m, 2H), 1.78 (m, 2H), 1.34-1.44 (m, 4H).

(1S,2S)-2-(4-methoxy-6-((2-(1-methyl-1H-1,2,3-triazol-4-ylamino)pyrimidin-4-yl) methoxy)benzo[d]thiazol-2-ylamino)cyclohexanol (31)

Step 1: (1S,2S)-2-(4-methoxy-6-((2-(1-methyl-1H-1,2,3-triazol-4-ylamino)pyrimidin-4-yl) methoxy)benzo[d]thiazol-2-ylamino)cyclohexanol. A mixture of (1S,2S)-2-(6-((2-chloropyrimidin-4-yl)methoxy)-4-methoxybenzo [d]thiazol-2-ylamino)cyclohexanol (60 mg, 0.14 mmol), 1-methyl-1H-1,2,3-triazol-4-amine (17 mg, 0.17 mmol), TsOH (12 mg, 0.07 mmol) in dioxane (2 ml) was stirred at 110°C for 16 h. The mixture was diluted with EtOAc, washed with $NaHCO_3$ (aq. Sat.) and dried over Na_2SO_4. The combined organic layer were concentrated under reduced pressure. The residue was purified by preparative HPLC (Mobile phase: A = 0.1% NH_4HCO_3/H_2O, B = MeCN; Gradient: B = 5 - 95%; 12 min; Column: C18) to give the title compound (7.0 mg, 10%) as a white solid. MS (ES+) $C_{22}H_{26}N_8O_3S$ requires: 482 found: 483 $[M+H]^+$. ^1H NMR (500 MHz, DMSO-d_6) δ 10.25 (s, 1H), 8.47 (d, *J* = 5 Hz, 1H), 8.13 (s, 1H), 7.61 (d, J = 7.5 Hz, 2H), 6.94-6.96 (m, 2 H), 6.60 (s, 1 H), 5.10 (s, 2H), 4.76 (d, *J* = 5 Hz, 1H), 4.00 (s, 3H), 3.83 (s, 3H), 2.02-2.05 (m, 1H), 1.86-1.88 (m, 1H), 1.60-1.65 (m, 2H), 1.18-1.30 (m, 4H).

Synthesis of (1S,2S)-2-((4-methoxy-6-((2-(pyridin-3-ylamino)pyrimidin-4-yl)methoxy)benzo[d]thiazol-2-yl)amino)cyclohexan-1-ol (32)

Step 1: (1S,2S)-2-((4-methoxy-6-((2-(pyridin-3-ylamino)pyrimidin-4-yl)methoxy)benzo[d]thiazol-2-yl)amino)cyclohexanol. A microwave vial was charged with (1S,2S)-2-((6-((2-chloropyrimidin-4-yl)methoxy)-4-methoxybenzo[d]thiazol-2-yl)amino)cyclohexanol (20 mg, 0.48 mmol), pyridin-3-amine (14 mg, 0.14 mmol), one drop of HCl (aq. 36.5%) and MeOH (1.5 mL). The vial was sealed and the reaction mixture was heated to 110 °C in the microwave reactor for 3 h. The residue was purified by mass-triggered preparative HPLC (Mobile phase: A = 0.1% TFA/H_2O, B = 0.1% TFA/MeCN; Gradient: B = 20 - 50%; 12 min; Column: C18) to give (1S,2S)-2-((4-methoxy-6-((2-(pyridin-3-ylamino)pyrimidin-4-yl)methoxy)benzo[d]thiazol-2-yl)amino)cyclohexanol (11 mg, 48%) as an off-white solid. MS (ES+) $C_{24}H_{26}N_6O_3S$ requires: 478.6, found: 479.5 [M+H]. 1H NMR (600 MHz, d_4-MeOD) δ 9.40 (s, 1H), 9.28 (m, 1H), 9.10 (d, J = 4.8 Hz, 1H), 8.02 (d, J = 4.8 Hz, 1H), 7.91 (m, 2H),7.07 (d, J = 2.4 Hz, 1H), 6.93 (d, J = 1.8 Hz, 1H), 5.44 (s, 2H), 4.02 (s, 3H), 3.60 (m, 1H), 3.47-3.51 (m, 1H), 2.05 – 2.1 (m, 2H), 1.79 (m, 2H), 1.34-1.46 (m, 4H).

Synthesis of (1S,2S)-2-(7-chloro-4-methoxy-6-((2-(1-methyl-1H-pyrazol-4-ylamino)pyrimidin-4-yl)methoxy)benzo[d]thiazol-2-ylamino)cyclohexanol (33)

Step 1: (1S,2S)-2-(7-chloro-6-((2-chloropyrimidin-4-yl)methoxy)-4-methoxybenzo[d]thiazol-2-ylamino)cyclohexanol: A mixture of 7-chloro-2-((1S,2S)-2-hydroxycyclohexylamino)-4-methoxybenzo[d]thiazol-6-ol (350 mg, 1.07 mmol), 2-chloro-4-(chloromethyl)pyrimidine (208 mg, 1.28 mmol) and Cs_2CO_3 (698 mg, 2.14 mmol) in DMF (8 ml) was stirred at 80°C for 3 h. The volatiles were removed under reduced pressure to give the residue, which was purified by column chromatography (PE: EA = 1:9) to give (1S,2S)-2-(7-chloro-6-((2-chloropyrimidin-4-yl)methoxy)-4-methoxybenzo[d]thiazol-2-ylamino)cyclohexanol (3) (185 mg, 35%) as a yellow solid. MS (ES+) $C_{19}H_{20}Cl_2N_4O_3S$ requires: 454 found: 455 $[M+H]^+$.

Step 2: (1S,2S)-2-(7-chloro-4-methoxy-6-((2-(1-methyl-1H-pyrazol-4-ylamino)pyrimidin-4-yl)methoxy)benzo[d]thiazol-2-ylamino)cyclohexanol. A mixture of (1S,2S)-2-(7-chloro-6-((2-chloropyrimidin-4-yl)methoxy)-4-methoxybenzo[d]thiazol-2-ylamino)cyclohexanol (3) (60 mg, 0.13 mmol), 1-methyl-1H-pyrazol-4-amine (21 mg, 0.15 mmol), TsOH (11 mg, 0.065 mmol) in dioxane (2 ml) was stirred at 110°C overnight. The

mixture was diluted with EtOAc, washed with $NaHCO_3$ (aq. sat.) and dried over Na_2SO_4. The combined organic layer were concentrated under reduced pressure. The residue was purified by preparative HPLC (Mobile phase: A = 0.1% NH_4HCO_3/H_2O, B = MeCN; Gradient: B = 5 - 95%; 12 min; Column: C18) to give (1S,2S)-2-(7-chloro-4-methoxy-6-((2-(1-methyl-1H-pyrazol-4-ylamino)pyrimidin-4-yl)methoxy)benzo[d]thiazol-2-ylamino)cyclohexanol (3.5 mg, 5%) as a white solid. MS (ES+) $C_{23}H_{26}ClN_7O_3S$ requires: 515 found: 516 $[M+H]^+$. ^1H NMR (500 MHz, DMSO-d_6) δ 9.51 (s, 1H), 8.44 (d, J = 4.5 Hz, 1H),7.89 (d, J = 9.5 Hz, 2H),7.48 (s, 1 H), 6.83-6.89 (m, 2 H), 5.17 (s, 2H), 4.17 (d, J = 5.5 Hz, 1H), 3.77 (s, 3H), 3.49 (s, 3H), 2.02-2.08 (m, 1H), 1.86-1.89 (m, 1H), 1.60-1.65 (m, 2H), 1.19-1.30 (m, 4H).

ASSOCIATED CONTENT

Supporting information

Assay conditions, materials and methods, kinase profiling data for compound **12** [Table S1] and for IACS-9439 (**1**) [Table S3] and a csv file containing molecular formula strings.

The Supporting Information is available free of charge on the ACS Publication website at DOI X.

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The manuscript was written through contributions of all authors. All authors have given approval to the final version of the manuscript.

Notes

The authors declare no competing financial interest.

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ABBREVIATIONS

°C, Celsius; ^{1}H-NMR, proton nuclear magnetic resonance; can, acetonitrile; AcOH, acetic acid; BSA, bovine serum albumin; BSTFA, *N*,*O*-Bis(trimethylsilyl)trifluoroacetamide; d_4-MeOD, deuterated methanol; DMSO-d_6, deuterated dimethyl sulfoxide; DBU, 1,8-diazabicycloundec-7-ene; DCM, dichloromethane; DIBAL, diisobutylaluminum hydride; DIEA, N,N-diisopropylethylamine; DMA, dimethyl acetamide; DMF, dimethylformamide; DMSO, dimethyl sulfoxide; DPPA, diphenylphosphoryl azide; DTT, dithiothreitol; ES^+, electrospray positive ionization; EGTA, ethylene glycol-bis(β-aminoethyl ether)-N,N,N',N'-tetraacetic acid; ELISA, enzyme-linked immunosorbent assay; Et_2O, diethyl ether; EtOAc, ethyl acetate; FRET, fluorescence resonance energy transfer; h, hour; HATU, 1-

[bis(dimethylamino)methylene]-1H-1,2,3-triazolo[4,5-b]pyridinium 3-oxide hexafluorophosphate; HEPES, 4-(2-hydroxyethyl)-1-piperazineethanesulfonic acid; HI FBS, heat inactivated fetal bovine serum; HPLC, high pressure liquid chromatography; HRP, horseradish peroxidase; Hz, hertz; IPA, isopropanol; M, molar; mCPBA, 3-chloroperbenzoic acid; MeCN, acetonitrile; MHz, megahertz; min, minute; mL, milliliter; MS, mass spectrometry; MW, microwave; PBS, phosphate buffer saline; PMB, p-methoxybenzyl; qPCR, quantitative polymerase chain reaction; rt, room temperature; Selectfluor, 1-Chloromethyl-4-fluoro-1,4-diazoniabicyclo[2.2.2]octane bis(tetrafluoroborate); tBuONO, tert-butyl nitrite; TFA, trifluoroacetic acid; THF, tetrahydrofuran.

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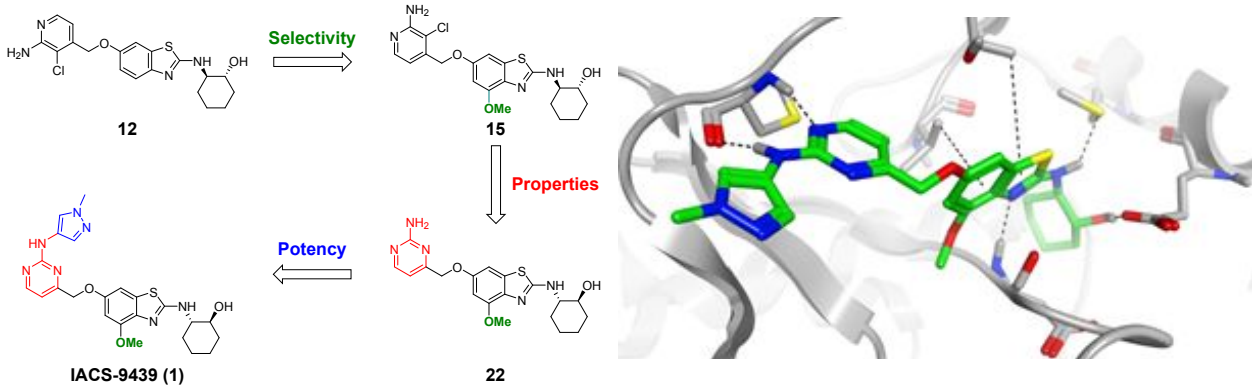
Table of Contents Graphic.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