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Discovery of a Diaminopyrimidine FLT3 Inhibitor Active against Acute Myeloid Leukemia

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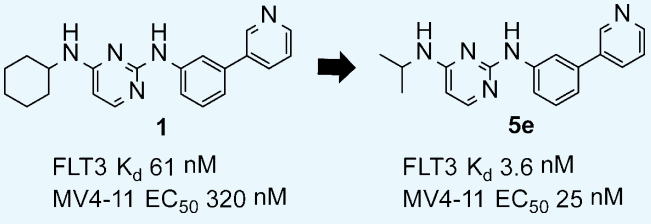
Supporting Information

ABSTRACT: Profiling of the kinase-binding capabilities of an aminopyrimidine analogue detected in a cellular screen of the St. Jude small-molecule collection led to the identification of a novel series of FMS-like tyrosine kinase 3 (FLT3) inhibitors. Structure−activity relationship studies led to the development of compounds exhibiting good potency against MV4-11 and MOLM13 acute myelogenous leukemia cells driven by FLT3, regardless of their FLT3 mutation status. In vitro pharmacological profiling demonstrated that compound **5e** shows characteristics suitable for further preclinical development.



INTRODUCTION

Acute myelogenous leukemia (AML) is characterized by malignant proliferation of hematopoietic cells in the bone marrow, leading to overproduction of abnormally functioning white blood cells. This situation leads to decreased production of red blood cells, infection, and dysfunction of organs. AML occurs in roughly 4 per 100 000 individuals; more than half of the reported cases occur in individuals over 65 years of age.[1] Although the use of hematopoietic stem cell transplants has provided some improved clinical outcomes in AML patients, there has been very little improvement in patient prognosis over the past 20 years.[2]

Normal functioning of FMS-like tyrosine kinase 3 (FLT3), a type III receptor tyrosine kinase, is important for the development and proliferation of hematopoietic stem cells.[3,4] The binding of the FLT3 ligand to this transmembrane protein causes dimerization and subsequent FLT3 autophosphorylation, which then triggers the activation of several signaling cascades, including the RAS, SRC, and STAT5 pathways.[5−8] Constitutive activation of FLT3 leads to dysregulated cellular proliferation of hematopoietic cells, and nearly one-third of AML patients have mutations in the FLT3 gene.[9] Two classes of mutations are commonly found in FLT3: an internal tandem duplication (ITD) located in the juxtamembrane domain, which is the most common, and point mutations at or near residue Asp835.[10−14] Typically, an AML patient's prognosis is worse if he/she possesses the FLT3-ITD mutation compared to that for patients having normal levels of wild-type FLT3 (wt-FLT3).[15,16]

Initially, kinase inhibitors developed for solid tumors were investigated as FLT3 inhibitors. Several of these inhibitors, including midostaurin,[17−19] lestaurtinib,[20−22] crenolanib,[23−26] tandutinib,[27−29] sunitinib,[30−32] and sorafenib,[33−36] have been evaluated in clinical trials. These compounds tended to inhibit multiple tyrosine kinases, thus leading to toxicity due to off-target effects.[37] Subsequently, quizartinib and crenolanib were developed as more selective FLT3 inhibitors.[38−42] The clinical response to FLT3 inhibitors often persists only for a short duration, with acquired point mutations in FLT3 that impact the binding of the inhibitors driving the limited response.[37,43,44] In particular, FLT3 Asp835 mutants tend to be resistant to "type II" kinase inhibitors that bind to an inactive conformation of the enzyme, wherein the inhibitor is able to make contacts within an allosteric pocket adjacent to the ATP site due to the Asp-Phe-Gly (DFG) motif in the activation loop adopting a conformation in which it is flipped out relative to its active conformation.[45−48] However, treatment with different FLT3 inhibitors can lead to alternate sets of acquired mutations, and some "type I" ATP-pocket-binding inhibitors, which bind within the ATP site but do not reach into the allosteric pocket and do not rely on specific DFG motif conformations, such as crenolanib, are able to bind selectively to FLT3 and also retain their activity against FLT3 Asp835 mutants.[23,49,50] Consequently, the development of additional new FLT3 inhibitors that can retain their activity against commonly acquired mutations or the use of FLT3 inhibitors in combination therapies may be potential methods to circumvent the problem of resistance.

While investigating potential compounds of interest identified during a cellular high-throughput phenotypic screen for a brain tumor project (results not yet published), the kinase-binding profile of one of the hits suggested potential use as a FLT3

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inhibitor. We therefore synthesized a series of compounds based on this hit for evaluation against FLT3 and investigation of their activities in AML cell lines. Herein we describe the structure−activity (SAR) and structure−property relationships resulting from this series of molecules.

RESULTS AND DISCUSSION

Preliminary Profiling of the Hit (1). The kinase-binding profile of **1** was evaluated using the DiscoveRx KINOMEscan panel of 468 kinases, with ligand competition being measured at a single inhibitor concentration of **1** (10 μM).[51−53] Selectivity was evaluated through a comparison of the number of nonmutant kinases with which **1** interacted relative to the total number of nonmutant kinases tested. Compound **1** reduced ligand binding by >90% for 31 kinases of 403 nonmutant kinases tested. Within that set, seven had activity reduced to >99% of that of the control. As depicted in Figure 1, **1** bound with the highest affinity to

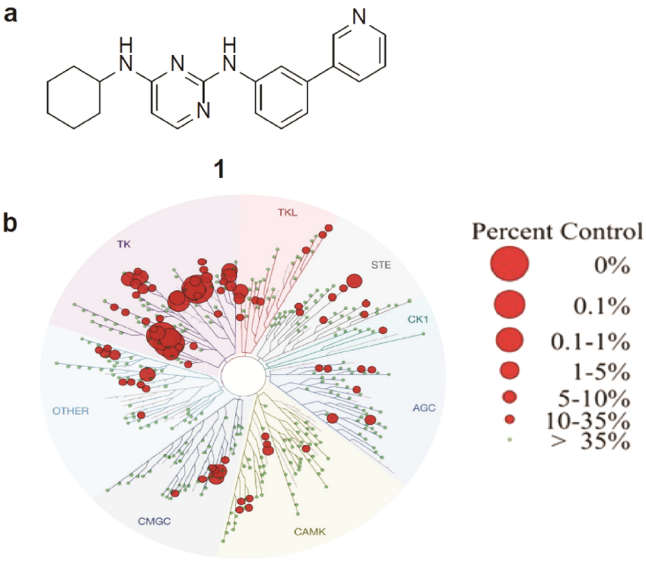

Figure 1. Lead series analogue **1**. (a) Structure of **1**. (b) Kinase profiling of **1**. Kinase tree image showing relative inhibition of ligand binding to 468 human kinases, with the size of the circle being proportionate to the % inhibition at a fixed dose of 10 μM.

kinases in the tyrosine kinase family. Further information on the kinase-binding profile is found in Table S1. Subsequently, the K_d values were determined for kinases in which compound **1** had blocked the binding of ligand by >90%, revealing that **1** bound with a high affinity to FLT3 (61 nM) and a significant but lower affinity to PDGFRB, the JAK1 JH2 domain, and JAK2 JH1 domain (Table 1). Eleven of the kinases had binding affinities within tenfold of those for the FLT3 primary target, which is a reasonably selective binding profile. For instance, previously studied kinase inhibitors, such as the AKT1 inhibitor GSK-690693, the IGF1R inhibitor GSK-1838705A, and the mammalian target of rapamycin inhibitor PP-242, had similar relative selectivity profiles.[54] We also verified that compound **1** inhibited the enzymatic activity of FLT3. As measured using the Z′LYTE kinase inhibition assay (ThermoFisher), **1** was a potent inhibitor of the FLT3^{WT} enzyme (IC_{50} = 32 nM; 95% confidence interval (CI 95), 24−43 nM).[55] Compound **1** was also a reasonably potent inhibitor of proliferation for the well-characterized MV4-11 AML cell line, which carries an FLT3-ITD mutation (EC_{50} = 320 nM; CI 95, 160−650 nM). This finding suggested that **1** acts on FLT3-ITD in these cells. Although aminopyrimidines have been well studied as kinase inhibitors, low-molecular-weight structures that include an aliphatic moiety, such as **1**, are not as widely reported and, to the best of our knowledge, have not been described specifically as FLT3 kinase inhibitors.[56,57] Taken together, the promising activity profile and fairly novel structural character of **1** prompted us to explore additional SARs.

Chemistry. We developed a general route for the synthesis of the majority of the compounds discussed herein (Scheme 1) on the basis of prior work on Aurora kinase inhibitors.[58] The route, which relied upon sequential additions of an aliphatic amine and various anilines to 2,4-dichloropyrimidine, facilitated systematic variation of the amine substituents at the 2- and 4-positions of the pyrimidine ring. Whereas the first substitution using an aliphatic amine typically proceeded facilely at either 0 °C or room temperature (rt), subsequent reactions with the anilines required increased temperature and catalytic HCl. Anilines were obtained either from commercial vendors or were synthesized via the Suzuki reaction or other known procedures (Schemes S1−S6).[59,60] To help define the minimal pharmacophore, we investigated the impact of removing nitrogen atoms from the scaffold. Pyridine analogues were synthesized by the routes shown in Schemes 2 and 3. These involved nucleophilic addition of cyclohexylamine to the halopyrimidine, followed by a palladium-catalyzed coupling reaction to install the 3-pyridinyl-3-ylaniline functional group. Schemes 4−7 depict the routes used to synthesize analogues in which the amine linkers at the 2- and/or 4-positions of the aminopyrimidine were replaced with *N*-methyl or oxygen. *N*-Methylcyclohexane was installed at the 2-position by treating with triethylamine (Scheme 4), whereas cyclohexanol was incorporated by reacting with 2,4-dichloropyrimidine in the presence of sodium hydride (Scheme 5). Substituents at the 4-position were installed using catalytic HCl and by refluxing in ethanol (EtOH) (Schemes 4−7). Overall, the routes proceeded with sufficient yield (3−85% yield), producing all targeted analogues for characterization, although, undoubtedly, further optimization could be beneficial for individual analogues. All compounds used for subsequent testing were purified to greater than 95% purity, as confirmed by the combination of liquid chromatography−mass spectrometry (LC−MS) and ^{1}H NMR studies.

SAR. We utilized the FLT3^{WT} enzyme assay to determine the potencies of all analogues of **1** made during this study (Table S2). In parallel, we tested the effects on the proliferation of MV4-11 cells (Table S2). We also measured the effects of these compounds on the proliferation of untransformed BJ fibroblasts to evaluate the dependency of cellular effects upon the presence of a driving FLT3 mutation (Table S3). Because cellular assays include fetal bovine serum within the media, there is a potential that the EC_{50} values could differ from the actual values due to plasma protein binding.[61] The solubility and permeability as well as the potential susceptibility to P-glycoprotein (PGP) efflux were assessed for selected compounds (Tables S4 and S5) to aid in interpreting the data and planning for structural modifications.

The initial focus of the project was defining the minimal pharmacophore for the inhibitors. Replacement of the nitrogen at the 1-position with a carbon atom (**9**) did not have a large impact on potency for kinase inhibition but completely ablated effects upon proliferation of MV4-11 cells (Table 2). Removal of the nitrogen at the 3-position (**13**) reduced the potency of kinase inhibition compared to that of **1**, while adding weak antiproliferative effects on untransformed BJ cells (Table S3).

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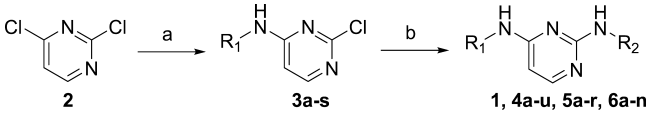
Table 1. K_d Values for 1

kinase	K_d (nM)[a]	kinase	K_d (nM)[a]
ABL1 (H396P)-phosphorylated	2100	LCK	2900
AURKA	1600	MERTK	2800
AURKC	>10 000	MINK	2500
AXL	2100	NEK10	410
BLK	3900	PDGFRB	120
CLK1	430	PIK4CB	370
CLK4	690	PIP5K1C	610
CSF1R	590	PRKCQ	1600
CSF1R-autoinhibited	1500	PRKD3	1700
EPHB6	560	RET (M918T)	1200
FLT3	61	RET (V804M)	1400
HCK	2100	RSK1 (Kin.Dom.1-N-terminal)	4400
IRAK1	1500	SRC	1100
IRAK3	1400	TRKA	1100
JAK1 (JH2 domain-pseudokinase)	290	TYK2 (JH1 domain-catalytic)	350
JAK2 (JH1 domain-catalytic)	260	TYK2 (JH2 domain-pseudokinase)	740
JAK3 (JH1 domain-catalytic)	1300	TYRO3	2700
KIT	1500		

[a]K_d values are reported as the mean of two experiments.

Scheme 1. General Route for the Synthesis of 2,4-Aminopyrimidine Analogues[a]



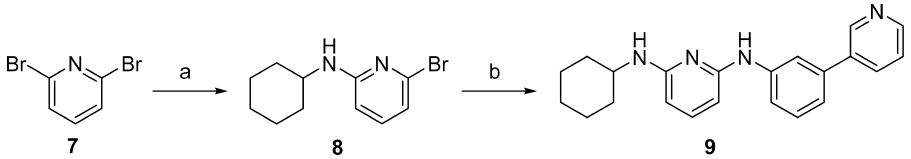
[a]Reagents and conditions: (a) NH_2-R_1, dichloromethane (DCM) or methanol (MeOH) or NH_2−R_1, triethylamine, DCM, rt, 5−16 h, 1−60%; (b) NH_2-R_2, catalytic HCl, EtOH or MeOH, reflux, 1−22 h, 3−85%.

Compound **14**, in which the location of the ring nitrogen had shifted, exhibited good enzymatic inhibitory potency but lacked MV4-11 cellular activity and also was weakly active against BJ fibroblasts (Table S3). Therefore, the pyrimidine core was necessary for optimal activity.

We examined the effects of replacing the linker atoms on both FLT3 inhibition and MV4-11 proliferation using both *N*-methyl and ether replacements (Table 3). Although replacement of the 2-amine with *N*-methyl abrogated activity against FLT3 (**21**, **17**), **17** possessed modest cellular potency, which might be due to metabolic removal of the methyl groups (unexplored). Replacement of the amine linkers with ethers at either the 4-position (**19**) or both the 2- and 4-position (**20**) did not greatly impact the enzymatic activity, whereas replacement of only the 2-nitrogen with an ether linkage did reduce the enzymatic activity nearly 40-fold compared to that of **1** (**22**). However, compounds **19**, **22**, and **20** were all devoid of effects on MV4-11 cell proliferation. Although the lack of enzymatic inhibition exhibited by N-methylated compounds **17** and **21** could suggest that a key hydrogen-bonding interaction may be made with FLT3 through the amine at the 2-position, compound **20**, which contains an ether at this location, also demonstrated potent FLT3 inhibition. Therefore, it is not clear what particular interactions occur between these compounds and the kinase. Overall, however, the inclusion of amine linkers at the 2- and 4-position provided the best combination of enzymatic and cellular activity. On the basis of these results, we focused on the 2,4-aminopyrimidine scaffold for further optimization.

Next, we explored the replacement of the pendent hydrophobic groups surrounding the aminopyrimidine scaffold. First, the cyclohexyl substituent at the 4-position was fixed, while the substituent at the 2-position was systematically varied (Table 4). Paring the structure down by removing the pyridine ring (**4a**) caused a reduction in FLT3 inhibitory activity and ablated activity in MV4-11 cells; therefore, this functional group may improve binding to FLT3 through a potential a H-bonding interaction. Reductions in FLT3 inhibition and MV4-11 cellular activity were also observed with compounds **4c−f**, which also did not contain the pyridine ring in **1**. Replacement of the 3-(pyridin-3-yl)aniline moiety with either 2- or 3-substituted biphenyl (**4b**, **4g**) abolished all activity. Incorporation of a 3′-methoxybiphenyl (**4i**) or methyl ester (**4j**) led to a reduction in enzymatic and MV4-11 cellular activities compared to those of **1**, and these compounds also gained weak activity against BJ cells (Table S3). Shifting the location of the ring nitrogen (**4h**) reduced the activity compared to that of **1**. However, replacement of the 3-

Scheme 2. Synthesis of Compound 9[a]



[a]Reagents and conditions: (a) aminocyclohexane, triethylamine, *N*-methyl-2-pyrrolidone (NMP), 100 °C, 21 h, 10%; (b) 3-pyridinyl-3-ylaniline, $Pd_2(dba)_3$, *R*-BINAP, sodium *tert*-butoxide, toluene, 80 °C, 16 h, 56%.

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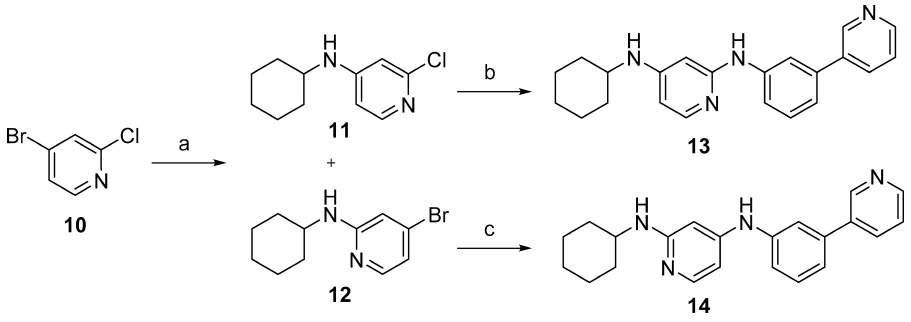
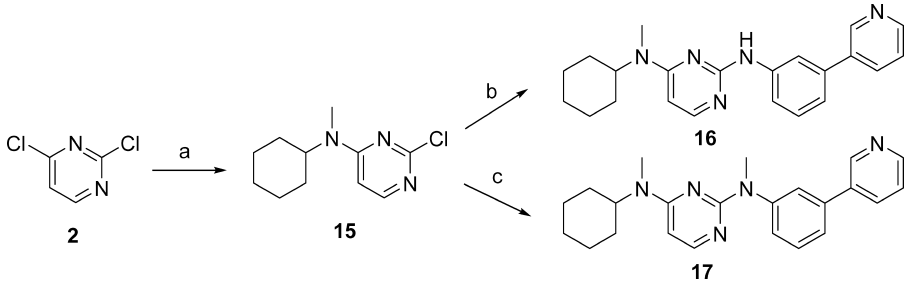
Scheme 3. Synthesis of Compounds 13 and 14[a]



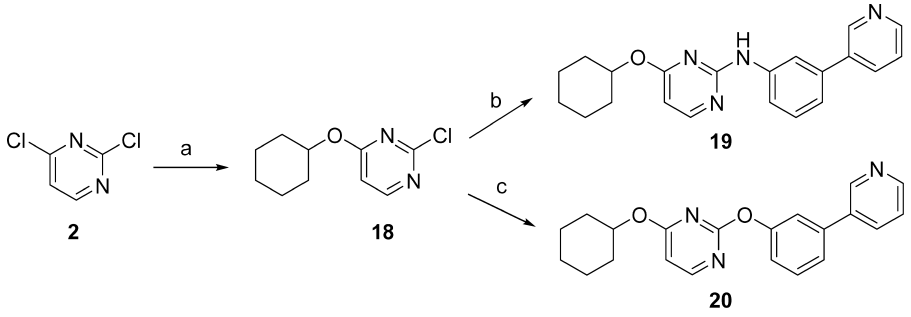
[a]Reagents and conditions: (a) aminocyclohexane, triethylamine, NMP, 80 °C, 16 h, 15% **11**, 2% **12**; (b) 3-pyridinyl-3-ylaniline, $Pd_2(dba)_3$, *R*-BINAP, sodium *tert*-butoxide, dimethylformamide (DMF), 140 °C, 35 mW, 1 h, 24%; (c) 3-pyridinyl-3-ylaniline, $Pd_2(dba)_3$, *R*-BINAP, sodium *tert*-butoxide, toluene, 80 °C, 16 h, 9%.

Scheme 4. Synthesis of Compounds 16 and 17[a]



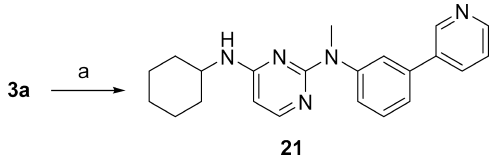
[a]Reagents and conditions: (a) *N*-methylaminocyclohexane, triethylamine, ethanol, reflux, 5 h, 59%; (b) 3-pyridinyl-3-ylaniline, catalytic HCl, ethanol, reflux, 3 h, 25%; (c) *N*-methyl-3-(pyridin-3-yl)aniline, catalytic HCl, ethanol, reflux, 2 h, 27%.

Scheme 5. Synthesis of Compounds 19 and 20[a]



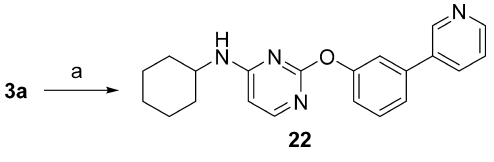
[a]Reagents and conditions: (a) cyclohexanol, sodium hydride, DMF, 0 °C to rt, 16 h, 3%; (b) 3-pyridinyl-3-ylaniline, catalytic HCl, ethanol, reflux, 3 h, 9%; (c) 3-(pyridin-3-yl)phenol, catalytic HCl, ethanol, reflux, 16 h, 52%.

Scheme 6. Synthesis of Compound 21[a]



[a]Reagents and conditions: *N*-methyl-3-(pyridin-3-yl)aniline, catalytic HCl, ethanol, reflux, 2 h, 6%.

Scheme 7. Synthesis of Compound 22[a]



[a]Reagents and conditions: 3-(pyridin-3-yl)phenol, catalytic HCl, ethanol, reflux, 16 h, 81%.

pyridinyl group with other nitrogen-containing heterocycles was tolerated by FLT3 (**4k**–**n**), further suggesting that a nitrogen atom in this position may provide critical interactions with FLT3. Compounds **4o**, **4p**, and **4r**, which also lacked a pyridinyl nitrogen, showed diminished FLT3 inhibition compared to that by **1** as well. Although compound **4q** contained the desired pyridine ring, altered molecular conformations may have led to this compound's lack of activity. Compound **4s** also exhibited good activity, suggesting that the carboxamide moiety may be in a beneficial location for hydrogen-bonding interactions with FLT3. Inclusion of a pyridine ring at the para position (**4t**) provided modest FLT3 activity but good MV4-11 cellular

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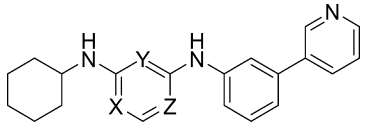
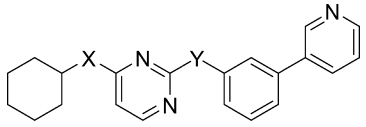
Table 2. SAR of the Aminopyrimidine Ring for Compounds 9, 13, 14, and Quizartinib



compd.	X	Y	Z	FLT3 IC_{50} (nM)[a]	MV4-11 EC_{50} (nM)[a]	BJ EC_{50} (nM)[a]
9	C	N	C	80	>10 000	>10 000
13	C	C	N	1500	>16 000	10 615
14	N	C	C	141	>8000	8741
quizartinib				40	0.7	>22 000

[a]IC_{50} and EC_{50} values are reported as the mean of triplicates. Values for CI 95 are included in Table S3.

Table 3. SAR Analysis of Modifications to the Linkers for Compounds 16, 17, and 19−22



compd.	X	Y	FLT3 IC_{50} (nM)[a]	MV4-11 EC_{50} (nM)[a]
16	N-Me	NH	82	>8000
21	NH	N-Me	>11 000	>4000
17	N-Me	N-Me	>15 000	39
19	O	NH	187	>6000
22	NH	O	1260	>14 000
20	O	O	49	>13 000

[a]IC_{50} and EC_{50} values are reported as the mean of triplicates. Values for CI 95 are included in Table S3.

activity. Although compound **4u** was inactive against FLT3, an effect on MV4-11 cell proliferation was observed. The potential off-target kinase activity of this compound was not investigated, but interactions with other kinases may contribute to the moderate cellular activity of compounds **4t** and **4u**. Taken together, these results point to a very strong preference for particular steric and electronic arrangements on this portion of the molecule, with the original 3-pyridinylphenyl presenting a good balance of properties.

In parallel, we synthesized a series of compounds keeping the 3-pyridinyl-3-ylaniline moiety constant, while varying the aliphatic substituents at the 4-position. Short linear, branched, and cyclic aliphatic groups were evaluated (Table 5). Although reduced enzymatic inhibition and a loss of MV4-11 cellular activity was observed when replacing the cyclohexyl group with a methyl (**5a**), good potency was obtained with either an ethyl (**5b**) or propyl (**5c**) group and moderate potency was obtained with a butyl (**5d**) group. Branched alkyl chains, such as isopropyl (**5e**), *sec*-butyl (**5j**, **5k**), and *tert*-butyl (**5f**), provided improvements in the FLT3 inhibitory activity and approximately 10-fold improvements in MV4-11 growth inhibition relative to those of **1**. Inclusion of ether (**5g**), ester (**5h**), or carboxylic acid (**5i**) functionalities within the branched aliphatic substituents greatly reduced cellular activity. Although **5g** had good solubility and permeability, the lower stability of the methoxy group in the cellular environment may have impacted its cellular activity (not explored). Some compounds with aliphatic rings, such as **5l** and **5m**, demonstrated good enzymatic and cellular activities. Although both the cyclopropyl and cyclobutyl (**5n**) groups afforded good potency against the FLT3 enzyme, poor solubility (Table S4) may have hampered the cellular activity of the latter. Despite its low solubility, compound **5o** exhibited good cellular activity. Therefore, additional properties beyond solubility could potentially have subtle effects on activity. Although **5q** was approximately 4-fold more potent than **1** against FLT3, the potency against MV4-11 cells was not improved due to poor cellular permeability (Table S4). Inclusion of a methoxy group (**5p**) was not beneficial. Replacing the amine functionality in the aliphatic ring of **5q** with an ether (**5r**) provided only moderate enzymatic and cellular activities.

Because of its improved activity against FLT3 and MV4-11 cells compared to that of **1**, we fixed the 4-position as the isopropyl group and varied the 2-position of the compound to determine whether the activity trends observed would be similar to those in the cyclohexyl series, as previously shown in Table 4. Similar to that in the cyclohexyl series, inclusion of a methoxy group (**6e**) or replacement of the pyridine functionality with a 2-, 3-, or 4-biphenyl (**6a**, **6c**, **6m**) caused decreases in both FLT3 inhibitory activity and cellular potency (Table 6). Inclusion of a *tert*-butyl group in place of the pyridine ring (**6b**) abrogated activity as well. Shifting the nitrogen from the 3-position to the 2-position (**6d**) or the 4-position (**6l**) also reduced potency, paralleling our prior observations and suggesting that a nitrogen at the 3-position may act as an H-bond acceptor to make an important binding interaction. Incorporation of a methyl ester (**6f**) led to reduced activity. The potency was comparable to that of **1** when the 3-pyridinyl group was replaced with a pyrazole (**6i**), and inhibition of MV4-11 cell proliferation improved when an imidazole (**6j**) or pyrazine (**6k**) replaced the pyridine. Compounds containing oxazole (**6g**) or thiazole (**6h**) moieties, however, provided less potent FLT3 inhibition and MV4-11 cellular activity. Similar to the effect observed with **4s**, the carboxamide-functionalized compound **6n** also exhibited moderate enzymatic inhibition and activity against MV4-11 cells.

On the basis of the balance of activity, we selected **5e** for further profiling. The compound retained its FLT3 potency (3.6 nM) and selectivity on the basis of profiling the K_d values for a subset of kinases inhibited by **1** (Table 7). Despite potent binding to PDGFRB (28 nM) and the JAK1 JH2 domain (28 nM), **5e** was still approximately 8-fold more potent against FLT3. Therefore, modifications explored during the SAR studies did not appear to significantly affect kinase selectivity compared to that of the initial hit compound.

In Vitro Effects against FLT3 Mutants and MOLM13 Cells. Because of the demonstrated activity of this series of compounds in MV4-11 cells, we evaluated a subset of compounds to determine whether they inhibited the proliferation of other FLT3 mutant AML cells. The initial hit compound **1** and **5e**, **6k**, and a structurally related inactive compound **6a** were tested for their ability to inhibit the proliferation of MOLM13 cells, which also contain the FLT3-ITD mutation, and a sorafenib-resistant progeny of MOLM13 that has acquired an additional tyrosine kinase domain (TKD) mutation, Asp835.[50] As shown in Figure 2, **6k** was the most potent at inhibiting MOLM13 cell viability (IC_{50} = 45 nM; CI 95, 38−55 nM), followed by **5e** (IC_{50} = 136 nM; CI 95, 112−164 nM). The relative sensitivity of MOLM13 cells to **5e** and **6k** was correlated to the loss of FLT3-ITD phosphorylation and downstream STAT5 signaling, with FLT3 IC_{50} values estimated to be within 50−100 nM in these experiments (Figure 3). In addition, all three tested compounds (**1**, **5e**, and **6k**) possessed similar growth inhibitory potencies against both parental $MOLM13^{FLT3\text{-}ITD}$ and

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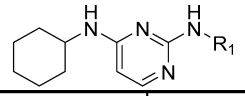
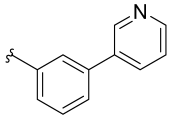
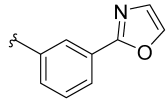
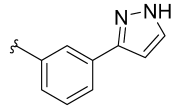
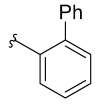
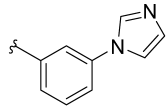
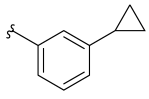
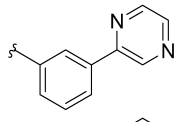
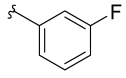
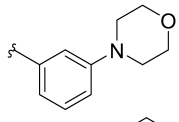
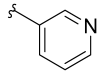
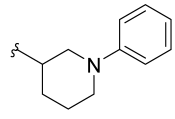
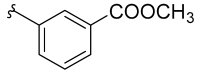
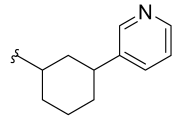
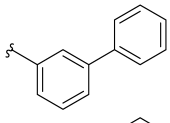
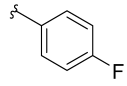
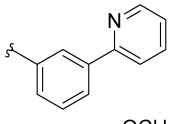
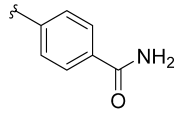
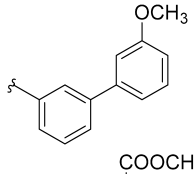
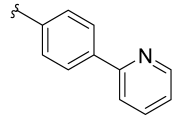
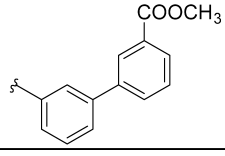
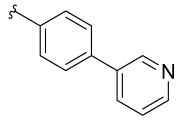
Table 4. SAR of the Substituents at the Aminopyrimidine 2-Position for Compounds 1 and 4a−u[a]



Compd.	R_1	FLT3 IC_{50} (nM)[a]	MV4-11 EC_{50} (nM)[a]	Compd.	R_1	FLT3 IC_{50} (nM)[a]	MV4-11 EC_{50} (nM)[a]
1		32	320	**4k**		336	511
4a		1890	>7000	**4l**		96	>14000
4b		>13000	>5000	**4m**		16	58
4c		2590	>15000	**4n**		20	53
4d		379	5050	**4o**		722	369
4e		>19000	>7000	**4p**		3260	3570
4f		978	5230	**4q**		>23000	>8000
4g		>12000	>4000	**4r**		3300	>3000
4h		1710	3230	**4s**		14	113
4i		24500	>15000	**4t**		1880	41
4j		13300	7590	**4u**		>31000	1260

[a]IC_{50} and EC_{50} values are reported as the mean of triplicates. Values for CI 95 are included in Table S3.

progeny MOLM13$^{FLT3\text{-}ITD/D835Y}$ cells. We also determined the K_d values for compounds **1**, **5e**, and **6a** against several FLT3 mutants (Table 8).[62] Compound **5e** bound more tightly to all FLT3 variants tested compared with **1**, whereas **6a** did not bind to any FLT3 variant at any tested concentration. Therefore, there was very good correlation between the improved potency of **5e** against MOLM13 cells and improved binding to FLT3 variants. Interestingly, the fold-difference in the affinities of **5e** and **1** was very similar between the FLT3 and Asp835 mutants, which is mirrored by similar potencies in parental and kinase inhibitor-resistant MOLM13 cells. This is a desirable profile for a new FLT3 inhibitor scaffold. Previously studied type I kinase inhibitors have also had similar to better affinities for FLT3, FLT3-ITD, or activating TKD mutations, suggesting that these

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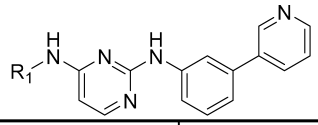
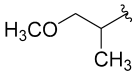
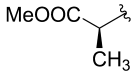
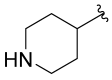
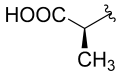
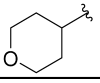
Table 5. SAR of the Substituents at the Aminopyrimidine 4-Position for Compounds 5a−r[a,b]



Compd.	R_1	FLT3 IC_{50} (nM)[a]	MV4-11 EC_{50} (nM)[a]	Compd.	R_1	FLT3 IC_{50} (nM)[a]	MV4-11 EC_{50} (nM)[a]
5a		240	>20000	**5j**		8	65
5b		47.5	35	**5k**		<6[b]	25
5c		<6[b]	138	**5l**		<6[b]	141
5d		123	340	**5m**		<6[b]	31
5e		<6[b]	25	**5n**		<6[b]	>17000
5f		13	25	**5o**		13	58
5g		<6[b]	>23000	**5p**		>34000	>12000
5h		>22000	>8000	**5q**		8	654
5i		>23000	>8000	**5r**		187	1877

[a]IC_{50} and EC_{50} values are reported as the mean of triplicates. Values for CI 95 are included in Table S3. [b]On the basis of assay conditions, IC_{50} values below 6 nM cannot be accurately measured.

compounds may have the properties of a type I inhibitor, which binds in the ATP-binding pocket of the kinase.[63] Additionally, when selected compounds were evaluated in the Z′LYTE FLT3 kinase inhibition assay under standard conditions but using either a lower (100 μM) or higher (2.5 mM) concentration of ATP, shifts in the FLT3 IC_{50} values of these compounds were observed. For instance, the IC_{50} of the initial hit, **1**, was reduced to <6 nM in the presence of 100 μM ATP and increased to 116 nM (CI 95, 70−193 nM) when the assay was conducted at a 2.5 mM ATP concentration. Similar to other type I kinase inhibitors, these compounds may make crucial binding interactions within the ATP site and therefore may not be sensitive to the conformation of the DFG motif.

In Vitro ADME Studies. We evaluated the in vitro pharmacological characteristics of **1**, **5e**, and **6k** to assess their potential for use in in vivo studies. All three analogues had excellent passive permeabilities ($1880 \pm 170 \times 10^{-6}$, $2000 \pm 190 \times 10^{-6}$, and $1870 \pm 60 \times 10^{-6}$ cm/s, respectively), as measured in the PAMPA assay. Neither **1** nor **5e** showed susceptibility to PGP efflux (Table S5) in cellular permeability assays. Compound **6k** was not evaluated in this assay. Although the solubility of **1** in PBS was poor (0.4 ± 0.9 μM), that of **5e** was reasonable (26.4 ± 17.0 μM). Finally, the stability of these three compounds in liver microsomes from four species was lower than desirable (Table 9). However, the plasma stability of **5e** and **6k** was excellent (>48 h). Overall, these studies would predict reasonable oral absorption for **5e** but rapid clearance (CL) for all three, and it would be expected that first-pass metabolism effects could cause low bioavailability.

In Vivo Pharmacokinetics. On the basis of the results from the microsomal stability studies of **1**, **5e**, and **6k**, we selected **5e** to evaluate in vivo pharmacokinetics at three doses by intraperitoneal (i.p.) injection. A greater than proportional dose-dependent increase in C_{max} (Table 10) was observed (327, 927, and 1625 nM for 3, 5, and 10 mg/kg respectively) and the half-life of the compound ranged from 0.23 to 1 h, paralleling our observations in mouse microsome models (0.2−1 h at 0.8−20 μM). The area under the concentration−time curve (AUC) within 24 h also displayed a dose-dependent trend. CL, however, was high at all doses tested. Therefore, because of the undesirable pharmacokinetic profile of **5e**, we did not pursue efficacy modeling using a human AML xenograft model with this compound because its pharmacokinetic properties could potentially confound the results of such a study.

CONCLUSIONS

Starting with hit compound **1**, we developed a SAR concerning inhibition of FLT3 enzymatic activity and MV4-11 cellular proliferation. Overall, the SAR for enzymatic inhibition and inhibition of cellular proliferation correlated well. We found that the pyrimidine nitrogens and nitrogen linker atoms were important for MV4-11 cellular activity. Inclusion of a 3-pyridinylphenyl group at the 2-position and a small aliphatic substituent at the 4-position provided a balance of potency, solubility, and permeability (Figure 4). Additionally, the pan-

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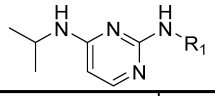
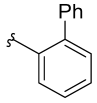
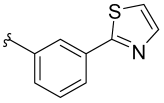
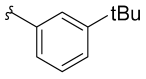
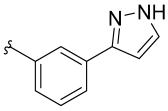
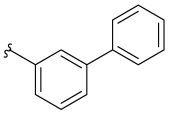
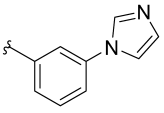
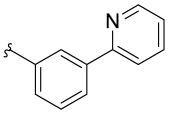
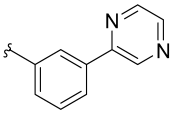
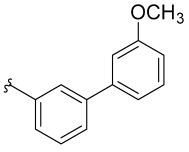
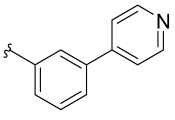
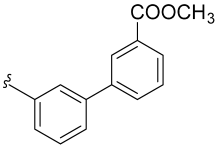
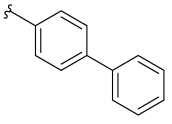
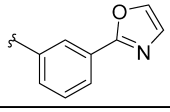
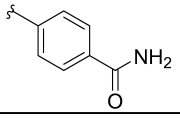
Table 6. Additional SAR at the Aminopyrimidine 2-Position for Compounds 6a−n[a,b]



Compd.	R_1	FLT3 IC_{50} (nM)[a]	MV4-11 EC_{50} (nM)[a]	Compd.	R_1	FLT3 IC_{50} (nM)[a]	MV4-11 EC_{50} (nM)[a]
6a		>24000	>16000	**6h**		475	9460
6b		17200	>13000	**6i**		42	459
6c		11600	3980	**6j**		<6[b]	4
6d		2880	4820	**6k**		11	5
6e		6970	>16000	**6l**		284	>15000
6f		4440	4890	**6m**		15000	11900
6g		918	1060	**6n**		213	2530

[a]IC_{50} and EC_{50} values are reported as the mean of triplicates. Values for CI 95 are included in Table S3. [b]On the basis of assay conditions, IC_{50} values below 6 nM cannot be accurately measured.

Table 7. K_d Values for 5e

kinase	K_d (nM)[a]
CLK1	260
CLK4	1100
CSF1R	460
EPHB6	440
FLT3	3.6
JAK1 (JH2 domain-pseudokinase)	28
JAK2 (JH1 domain-catalytic)	480
NEK10	2000
PDGFRB	28
PIK4CB	330
PIP5K1C	1300
TYK2 (JH1 domain-catalytic)	740
TYK2 (JH2 domain-pseudokinase)	210

[a]K_d values are reported as the mean of two experiments.

activity against FLT3 mutations and the relatively high selectivity for FLT3 were both generally maintained during the SAR studies, suggesting that the scaffold might provide good lead molecules for further development. From these studies, we developed **5e**, which not only had improved potency against FLT3 and MV4-11 cells but also demonstrated activity in MOLM13 cells possessing FLT3-ITD and FLT3 inhibitor-resistant TKD mutations. Compound **5e** possessed acceptable in vitro ADME properties and was selected for additional preclinical studies. However, because of its low metabolic stability and high CL in pharmacokinetic studies, we determined that these properties need to be improved before further preclinical studies are undertaken. Efforts toward this goal are on-going.

EXPERIMENTAL SECTION

Chemistry. All chemical reagents were purchased from commercial suppliers (Acros, Combi-Blocks, Enamine, Oakwood, Sigma Aldrich, Strem) and were used without further purification. Solvents were dried using a column-exchange system[64] Thin-layer chromatography (TLC) was performed using Merck Millipore silica gel 60G F_{254} glass plates and visualized using a 254 nm UV lamp for detection. Microwave

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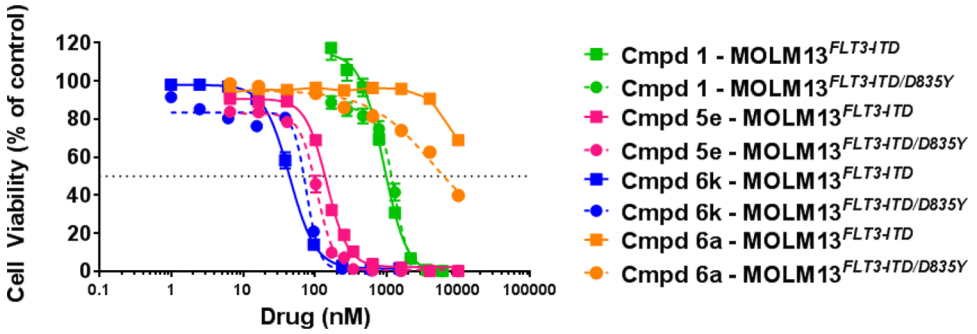

Figure 2. Relative inhibition of the proliferation of MOLM13 cells carrying either FLT3-ITD or FLT3-ITD with an additional mutation of D835Y by compounds **1**, **5e**, **6k**, and **6a**, as evaluated in the MTT assay. Data are expressed as the mean of cell viability measurements ± SE of three experiments, with six replicates each (n = 18), after treatment with drug for 72 h. (IC_{50} and CI 95 in nM: MOLM13$^{FLT3\text{-}ITD}$_**1** = 620, 461−833; MOLM13$^{FLT3\text{-}ITD/D835Y}$_**1** = 1153, 869−1604; MOLM13$^{FLT3\text{-}ITD}$_**5e** = 136, 112−164; MOLM13$^{FLT3\text{-}ITD/D835Y}$_**5e** = 82, 66−103; MOLM13$^{FLT3\text{-}ITD}$_**6k** = 45, 38−55; MOLM13$^{FLT3\text{-}ITD/D835Y}$_**6k** = 77, 63−94; MOLM13$^{FLT3\text{-}ITD}$_**6a** = >10 000; MOLM13$^{FLT3\text{-}ITD/D835Y}$_**6a** = 4260, 3060−5930.)

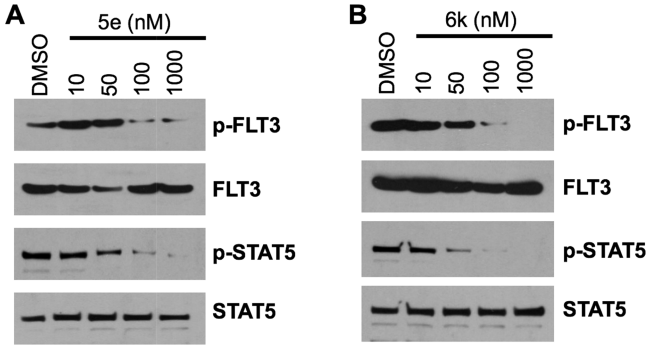

Figure 3. Inhibition of FLT3 signaling by compounds **5e** and **6k**. MOLM13$^{FLT3\text{-}ITD}$ cells were treated for 1 h with dimethyl sulfoxide (DMSO) or increasing concentrations of (A) compound **5e** or (B) compound **6k** and lysed. Western blot analysis was performed on the FLT3 immunoprecipitation eluent or the whole-cell lysate using the indicated antibodies.

Table 8. K_d Values for 1, 5e, and 6a Bound to Mutant FLT3 Kinases

kinase	**1** K_d (nM)[a]	**5e** K_d (nM)[a]	**6a** K_d (nM)[a]
FLT3	61	3.6	>10 000
FLT3 (D835H)	22	1.2	>10 000
FLT3 (D835V)	7.7	0.8	>10 000
FLT3 (D835Y)	13	0.59	>10 000
FLT3 (ITD)	31	1.4	>10 000
FLT3 (ITD, D835V)	7.2	0.36	>10 000
FLT3 (ITD, F691L)	270	11	>10 000
FLT3 (K663Q)	110	13	>10 000
FLT3 (N841I)	48	11	>10 000
FLT3 (R834Q)	680	82	>10 000
FLT3-autoinhibited	1100	78	>10 000

[a] K_d values are reported as the mean of two experiments.

experiments were carried out using a Biotage Initiator system. Automated flash chromatography was performed using the Biotage SP1 flash column system with silica gel SNAP columns or C18 SNAP columns for reversed-phase purifications.[65] R_f values are quoted for the eluent system stated. Evaporation was carried out using a Büchi Rotovapor. NMR spectra were recorded on either a Bruker 400 MHz or Bruker 500 MHz spectrometer in the solvents indicated, and the spectra were processed using MestReNova (8.1) referenced to the solvent peak. Melting points were obtained using a Büchi-545. Optical rotations were recorded using a Jasco P-1010 polarimeter at the D line of sodium (λ, 589 nM). Purity was assessed using ultra-performance liquid chromatography mass spectrometry (UPLC−MS; Acquity PDA detector, Acquity SQ detector, and Acquity UPLC BEH-C18 column, 1.7 μm, 2.1 × 50 mm^2; Waters Corp.). Data were acquired using Masslynx, version 4.1. The flow rate was 0.5 mL/min, and the gradient started with 95% A (0.1% formic acid in H_2O), changed to 95% B (0.1% formic acid in acetonitrile), and then returned to 95% A. The mass spectrometer was operated in the positive-ion mode with electrospray ionization. Verification of enantiopurity was conducted using supercritical fluid chromatography with a Chiralcel OD-H (4.6 × 250 mm^2) or Chiralcel AD-H (4.6 × 250 mm^2) column. The compounds were purified to ≥95% purity and assessed using LC/MS by UV/evaporative light scattering detector detection, unless stated otherwise, and efficacy data were obtained only on compounds that were ≥95% pure.

*2-Chloro-N-cyclohexylpyrimidin-4-amine (**3a**).* To a suspension of 2,4-dichloropyrimidine **2** (10 mmol, 1.0 equiv) in DCM (20 mL, 0.50 M) under a nitrogen atmosphere was added cyclohexylamine (20 mmol, 2.0 equiv) at rt. After stirring for 16 h at rt, the reaction mixture was concentrated and purified directly using automated flash chromatography (ethyl acetate (EtOAc)/hexanes), followed by evaporation, giving **3a** as a white solid (1.2 g, 57%). TLC R_f 0.30 (30% EtOAc/hexanes). LC−MS (ESI) m/z: 214 $[M + H]^+$. ^{1}H NMR (500 MHz, $CDCl_3$) δ 8.02 (s, 1H), 6.22 (d, J = 6.0 Hz, 1H), 5.20 (br s, 1H), 3.42 (br s, 1H), 2.08−1.93 (m, 2H), 1.77 (dt, J = 13.6, 4.0 Hz, 2H), 1.67 (dt, J = 12.9, 4.1 Hz, 1H), 1.38−1.44 (m, 2H), 1.32−1.13 (m, 3H). ^{13}C NMR (126 MHz, $CDCl_3$) δ 162.76, 160.83, 157.41, 100.54, 50.12, 32.68, 25.37, 24.51.

*N^4-Cyclohexyl-N^2-(3-(pyridin-3-yl)phenyl)pyrimidine-2,4-diamine (**1**).* To a mixture of **3a** (0.10 mmol, 1.0 equiv) in EtOH (0.50 mL, 0.20 M) under a nitrogen atmosphere at rt were added 3-(pyridin-3-yl)aniline (0.11 mmol, 1.1 equiv) and a drop of 1 N HCl. The reaction mixture was heated to reflux and stirred at that temperature for 3 h; it was then cooled to rt. Purification using automated flash chromatography (MeOH/DCM) was followed by evaporation, giving **1** as a white film (0.002 g, 6%). TLC R_f 0.5 (10% MeOH/DCM). LC−MS (ESI) m/z: 346 $[M + H]^+$. ^{1}H NMR (500 MHz, MeOD) δ 8.90−8.75 (s, 1H), 8.54 (d, J = 5.0 Hz, 1H), 8.11 (dt, J = 7.8, 2.0 Hz, 2H), 7.73 (s, 1H), 7.64 (d, J = 10 Hz, 1H), 7.54 (dd, J = 7.9, 4.9 Hz, 1H), 7.41 (t, J = 7.9 Hz, 1H), 7.25 (d, J = 10 Hz, 1H), 5.95 (d, J = 6.0 Hz, 1H), 3.84 (br s,

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Table 9. Physicochemical Properties of 1, 5e, and 6k

in vitro properties		1	5e	6k
solubility (μM)[a]	pH = 7.4	0.4 ± 0.9	26.4 ± 17.0	56.9 ± 1.9
permeability (10^{-6} cm/s)[a]		1880 ± 170	2000 ± 190	1870 ± 60
liver microsome stability at 4 μM $t_{1/2}$ (h)[a]	mouse	0.26 ± 0.03	0.30 ± 0.03	0.118 ± 0.003
	rat	1.04 ± 0.03	1.1 ± 0.1	0.38 ± 0.01
	dog	0.7 ± 0.1	1.2 ± 0.1	0.90 ± 0.02
	human	0.8 ± 0.1	2.1 ± 0.1	1.46 ± 0.10
liver microsome stability Clint at 4 μM (mL/min/kg)[a]	mouse	26.5 ± 1.6	179.6 ± 19.4	485.3
	rat	44.9 ± 1.4	41.7 ± 2.7	121.98
	dog	49.4 ± 3.9	28.2 ± 1.9	37.03
	human	26.6 ± 2.2	10.0 ± 0.6	14.22
plasma stability (h)[a]	mouse	N.D.[b]	>48	>50
	rat	N.D.[b]	>48	>50
	human	N.D.[b]	>48	>50
PBS stability (h)[a]	pH = 7.4	N.D.[b]	>48	N.D.[b]
	pH = 5.0	N.D.[b]	>48	N.D.[b]
	pH = 3.0	N.D.[b]	>48	N.D.[b]
SGF stability (h)[a]		N.D.[b]	>48	N.D.[b]

[a]Values are the mean of a single triplicate experiment. [b]N.D. means not determined.

Table 10. Pharmacokinetic Parameters in Mice for Compound 5e (i.p. Dosing)

dose (mg/kg)	3	5	10
C_{max} (nM)	376	927	1650
T_{max} (h)	0.083	0.083	0.25
$T_{1/2}$ (h)	0.23	0.54	1
AUC (nM h)	183.3	683.1	953.3
V_z (L/kg)	16.7	18.6	47.3
Cl (L/h/kg)	50.7	23.8	33.1

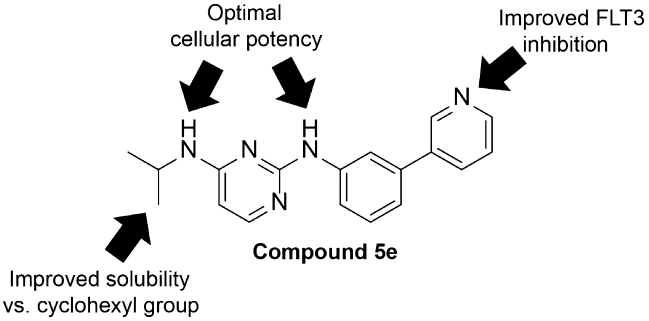

Figure 4. Summary of SAR.

1H), 1.98 (dd, J = 9.4, 4.8 Hz, 2H), 1.67−1.69 (m, 2H), 1.56 (s, 1H), 1.28−1.06 (m, 5H). ^{13}C NMR (126 MHz, MeOD) δ 162.32, 159.48, 153.10, 147.23, 147.10, 141.42, 137.93, 137.65, 135.32, 128.98, 124.06, 120.04, 119.00, 117.79, 97.65, 49.23, 32.43, 25.34, 24.72.

*6-Bromo-N-cyclohexylpyridin-2-amine (**8**).* A mixture of 2,6-dibromopyridine **7** (1.0 mmol, 1.0 equiv), cyclohexylamine (1.6 mmol, 1.6 equiv), and triethylamine (1.2 mmol, 1.2 equiv) was stirred at 100 °C in NMP (1.0 mL, 1.0 M) under a nitrogen atmosphere for 21 h; it was then cooled to rt. The reaction mixture was partitioned between EtOAc (3 mL) and water (3 mL). The organic phase was washed with brine (3 mL), dried over magnesium sulfate, filtered, and concentrated. Purification using automated flash chromatography (EtOAc/hexanes) was followed by evaporation, giving **8** as a colorless oil (0.026 g, 10%). TLC R_f 0.50 (10% EtOAc/hexanes). LC−MS (ESI) m/z: 257 $[M + H]^+$. ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.22−7.07 (m, 1H), 6.60 (d, J = 7.4 Hz, 1H), 6.18 (d, J = 8.2 Hz, 1H), 4.64−4.47 (br s, 1H), 3.41−3.26 (m, 1H), 1.89−1.96 (m, 2H), 1.72−1.61 (m, 2H), 1.53−1.59 (m, 1H), 1.39−1.23 (m, 2H), 1.23−1.05 (m, 3H). ^{13}C NMR (101 MHz, $CDCl_3$) δ 158.14, 140.32, 139.48, 115.22, 104.14, 50.36, 32.99, 25.66, 24.69.

*N^2-Cyclohexyl-N^6-(3-(pyridin-3-yl)phenyl)pyridine-2,6-diamine (**9**).* A mixture of **8** (0.078 mmol, 1.0 equiv), 3-(pyridin-3-yl)aniline (0.10 mmol, 1.3 equiv), sodium *t*-butoxide (0.39 mmol, 5.0 equiv), $Pd_2(dba)_3$ (0.005 mmol, 7 mol %), and *R*-BINAP (0.012 mmol, 15 mol %) was stirred in toluene (1.0 mL, 0.078 M) at 80 °C under a nitrogen atmosphere for 16 h; it was then cooled to rt. The reaction mixture was then diluted with EtOAc (3 mL) and washed with brine (2 × 3 mL). The organic phase was dried over magnesium sulfate, filtered, and concentrated. Purification using automated flash chromatography (EtOAc/hexanes) was followed by evaporation, giving **9** as a yellow oil (0.015 g, 56%). TLC R_f 0.70 (80% EtOAc/hexanes). LC−MS (ESI) m/z: 345 $[M + H]^+$. ^{1}H NMR (500 MHz, MeOD) δ 8.79 (s, 1H), 8.51 (s, 1H), 8.13−7.96 (m, 2H), 7.60 (d, J = 8.4 Hz, 1H), 7.58−7.48 (m, 1H), 7.34 (t, J = 8.0 Hz, 1H), 7.26−7.18 (m, 1H), 7.11 (d, J = 7.7 Hz, 1H), 6.02 (d, J = 8.1 Hz, 1H), 5.90 (d, J = 8.0 Hz, 1H), 3.77−3.59 (m, 1H), 2.11−1.93 (m, 2H), 1.75−1.59 (m, 2H), 1.54−1.68 (m, 1H), 1.15−1.22 (m, 5H). ^{13}C NMR (126 MHz, MeOD) δ 157.57, 157.53, 154.95, 154.91, 147.07, 143.20, 138.20, 138.10, 138.00, 137.89, 137.66, 135.29, 129.00, 124.03, 118.50, 117.77, 116.36, 97.90, 97.88, 96.85, 96.83, 49.80, 33.06, 25.60, 24.87.

*2-Chloro-N-cyclohexylpyridin-4-amine (**11**).* A mixture of 4-bromo-2-chloropyridine **10** (4.0 mmol, 1.0 equiv), cyclohexylamine (6.4 mmol, 1.6 equiv), and triethylamine (4.8 mmol, 1.2 equiv) was stirred at 80 °C in NMP (2.0 mL, 2.0 M) under a nitrogen atmosphere for 16 h. The reaction mixture was cooled to rt and then partitioned between EtOAc (3 mL) and water (3 mL). The organic phase was washed with brine (3 mL), dried over magnesium sulfate, filtered, and concentrated. Purification using automated flash chromatography (EtOAc/hexanes) was followed by evaporation, giving **11** as an orange solid (0.123 g, 15%). TLC R_f 0.40 (30% EtOAc/hexanes). LC−MS (ESI) m/z: 211 $[M + H]^+$. ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.92 (d, J = 5.8 Hz, 1H), 6.43 (s, 1H), 6.33 (dd, J = 5.8, 2.2 Hz, 1H), 4.26 (d, J = 7.8 Hz, 1H), 3.34−3.22 (m, 1H), 2.13−1.94 (m, 2H), 1.88−1.74

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(m, 2H), 1.74−1.61 (m, 1H), 1.47−1.32 (m, 2H), 1.32−1.12 (m, 3H). ^{13}C NMR (101 MHz, $CDCl_3$) δ 154.37, 152.16, 149.16, 107.41, 106.04, 51.03, 32.81, 25.54, 24.72.

*N^4-cyclohexyl-N^2-(3-(pyridin-3-yl)phenyl)pyridine-2,4-diamine (**13**).* A mixture of **11** (0.20 mmol, 1.0 equiv), 3-(pyridin-3-yl)aniline (0.25 mmol, 1.3 equiv), sodium *t*-butoxide (0.97 mmol, 5.0 equiv), $Pd_2(dba)_3$ (0.014 mmol, 7 mol %), and *R*-BINAP (0.029 mmol, 0.15 mmol) was stirred in DMF (1.0 mL, 0.20 M) at 140 °C under microwave irradiation at 35 W for 1 h. The reaction mixture was then cooled to rt, diluted with EtOAc (5 mL), and washed with brine (5 mL). The organic phase was dried over magnesium sulfate, filtered, and concentrated. Purification using automated flash chromatography (MeOH/CH_2Cl_2) was followed by evaporation, giving **13** as a brown oil (0.016 g, 24%). TLC R_f 0.3 (10% MeOH/CH_2Cl_2). LC−MS (ESI) *m*/*z*: 345 $[M + H]^+$. ^{1}H NMR (400 MHz, $CDCl_3$) δ 8.81 (s, 1H), 8.61−8.56 (m, 1H), 7.86 (dt, *J* = 7.9, 2.3 Hz, 1H), 7.57−7.50 (m, 1H), 7.48−7.42 (m, 2H), 7.35−7.38 (m, 1H), 7.27−7.32 (m, 2H), 6.12 (dd, *J* = 6.6, 2.1 Hz, 1H), 6.02 (s, 1H), 5.21 (d, *J* = 7.6 Hz, 1H), 3.27−3.14 (m, 1H), 1.92−1.97 (m, 2H), 1.76−1.68 (m, 2H), 1.59−1.61 (m, 1H), 1.16−1.30 (m, 5H). ^{13}C NMR (101 MHz, $CDCl_3$) δ 155.68, 153.72, 148.79, 148.15, 140.43, 139.34, 139.26, 135.98, 134.40, 130.21, 123.64, 123.14, 121.73, 120.74, 102.29, 87.65, 51.48, 32.59, 25.36, 24.58.

*4-Bromo-N-cyclohexylpyridin-2-amine (**12**).* A mixture of 4-bromo-2-chloropyridine **10** (1.0 mmol, 1.0 equiv), cyclohexylamine (1.6 mmol, 1.6 equiv), and triethylamine (1.2 mmol, 1.2 equiv) was stirred at 80 °C in NMP (1.0 mL, 1.0 M) under a nitrogen atmosphere for 16 h. The reaction mixture was cooled to rt and then partitioned between EtOAc (3 mL) and water (3 mL). The organic phase was washed with brine (3 mL), dried over magnesium sulfate, filtered, and concentrated. Purification using automated flash chromatography (EtOAc/hexanes) was followed by evaporation, giving **12** as a colorless oil (0.006 g, 2%). TLC R_f 0.75 (30% EtOAc/hexanes). LC−MS (ESI) *m*/*z*: 255 $[M + H]^+$. ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.88 (d, *J* = 5.5 Hz, 1H), 6.69 (dd, *J* = 5.5, 1.6 Hz, 1H), 6.55 (d, *J* = 1.6 Hz, 1H), 4.64 (s, 1H), 3.59−3.42 (m, 1H), 2.08−1.98 (m, 2H), 1.85−1.71 (m, 2H), 1.67 (dt, *J* = 12.7, 3.9 Hz, 1H), 1.53−1.33 (m, 2H), 1.33−1.14 (m, 3H). ^{13}C NMR (101 MHz, $CDCl_3$) δ 158.62, 148.77, 133.60, 115.61, 109.24, 50.28, 33.17, 25.68, 24.79.

*N^2-Cyclohexyl-N^4-(3-(pyridin-3-yl)phenyl)pyridine-2,4-diamine (**14**).* A mixture of **12** (0.067 mmol, 1.0 equiv), 3-(pyridin-3-yl)aniline (0.087 mmol, 1.3 equiv), sodium *t*-butoxide (0.33 mmol, 5.0 equiv), $Pd_2(dba)_3$ (0.0047 mmol, 7 mol %), and *R*-BINAP (0.010 mmol, 15 mol %) was stirred in toluene (1.0 mL, 0.067 M) at 80 °C under a nitrogen atmosphere. After 16 h, the reaction mixture was cooled to rt, diluted with EtOAc (3 mL), and washed with brine (2 × 3 mL). The organic phase was dried over magnesium sulfate, filtered, and concentrated. Purification using automated flash chromatography (MeOH/DCM) was followed by evaporation, giving **14** as a white oil (0.002 g, 9%). TLC R_f 0.2 (10% MeOH/DCM). LC−MS (ESI) *m*/*z*: 345 $[M + H]^+$. ^{1}H NMR (500 MHz, MeOD) δ 8.84 (s, 1H), 8.58 (s, 1H), 8.14 (d, *J* = 5.0 Hz, 1H), 7.68−7.46 (m, 5H), 7.39−7.30 (d, *J* = 5.0 Hz, 1H), 6.44 (dd, *J* = 7.0, 2.2 Hz, 1H), 6.25 (s, 1H), 3.40−3.44 (m, 1H), 1.99 (d, *J* = 15.0 Hz, 2H), 1.77−1.83 (m, 2H), 1.68 (dt, *J* = 12.8, 3.8 Hz, 1H), 1.37−1.46 (m, 2H), 1.24−1.34 (m, 3H). ^{13}C NMR (126 MHz, MeOD) δ 154.89, 153.53, 147.83, 146.98, 139.59, 138.87, 136.95, 136.53, 135.22, 130.27, 124.16, 123.63, 122.39, 121.05, 102.81, 88.10, 50.49, 32.29, 25.05, 24.37.

*2-Chloro-N-cyclohexyl-N-methylpyrimidin-4-amine (**15**).* A mixture of 2,4-dichloropyrimidine **2** (5.0 mmol, 1.0 equiv), *N*-methylaminocyclohexane (5.0 mmol, 1.0 equiv), and triethylamine (5.5 mmol, 1.1 equiv) was stirred in EtOH (7.0 mL, 0.71 M) under a nitrogen atmosphere at reflux temperature for 5 h. The reaction mixture was then cooled to rt and concentrated. Purification using automated flash chromatography (EtOAc/hexanes) was followed by evaporation, giving **15** as a white solid (0.67 g, 59%). TLC R_f 0.50 (30% EtOAc/hexanes). LC−MS (ESI) *m*/*z*: 228 $[M + H]^+$. ^{1}H NMR (500 MHz, $CDCl_3$) δ 8.00 (d, *J* = 6.2 Hz, 1H), 6.46−6.17 (m, 1H), 4.72 (br s, 1H), 2.93 (br s, 3H), 1.97−1.80 (m, 2H), 1.83−1.61 (m, 3H), 1.43−1.52 (m, 4H), 1.23−1.05 (m, 1H). ^{13}C NMR (126 MHz, $CDCl_3$) δ 162.58, 160.52, 156.66, 101.31, 29.82, 25.63, 25.48.

*N^4-Cyclohexyl-N^4-methyl-N^2-(3-(pyridin-3-yl)phenyl)-pyrimidine-2,4-diamine (**16**).* To a mixture of **15** (0.10 mmol, 1.0 equiv) in EtOH (0.50 mL, 0.20 M) under a nitrogen atmosphere at rt were added 3-(3-pyridyl)aniline (0.11 mmol, 1.1 equiv) and a drop of 1 N HCl. The reaction mixture was heated to reflux and stirred at that temperature for 3 h; it was then cooled to rt. Purification using automated flash chromatography (EtOAc/hexanes) was followed by evaporation, giving **16** as a colorless oil (0.009 g, 25%). TLC R_f 0.1 (70% EtOAc/hexanes). LC−MS (ESI) *m*/*z*: 360 $[M + H]^+$. ^{1}H NMR (500 MHz, $CDCl_3$) δ 8.86 (d, *J* = 2.3 Hz, 1H), 8.58 (dd, *J* = 4.8, 1.6 Hz, 1H), 7.94−7.84 (m, 2H), 7.61 (d, *J* = 8.7 Hz, 2H), 7.40 (t, *J* = 7.9 Hz, 1H), 7.35 (dd, *J* = 7.9, 4.8 Hz, 1H), 7.19 (d, *J* = 5.0, 1H), 5.98 (d, *J* = 6.3 Hz, 1H), 4.28 (br s, 2H), 2.92 (s, 3H), 1.84−1.72 (m, 2H), 1.70 (d, *J* = 3.9 Hz, 2H), 1.67−1.56 (m, 1H), 1.45 (q, *J* = 11.9, 11.1 Hz, 2H), 1.19−1.25 (m, 2H), 1.14−1.00 (m, 1H). ^{13}C NMR (126 MHz, $CDCl_3$) δ 162.03, 158.45, 154.29, 148.46, 148.44, 140.76, 138.53, 137.01, 134.49, 129.36, 123.47, 120.79, 118.85, 117.89, 95.37, 54.65, 30.01, 29.47, 25.75, 25.55.

*N^4-Cyclohexyl-N^2-methyl-N^2-(3-(pyridin-3-yl)phenyl)-pyrimidine-2,4-diamine (**21**).* To a mixture of **3a** (0.10 mmol, 1.0 equiv) in EtOH (0.50 mL, 0.20 M) under a nitrogen atmosphere at rt were added *N*-methyl-3-(pyridin-3-yl)aniline (0.11 mmol, 1.1 equiv) and a drop of 1 N HCl. The reaction mixture was heated to reflux and stirred at that temperature for 2 h; it was then cooled to rt. Purification using automated flash chromatography (MeOH/DCM) was followed by evaporation, giving **21** as a red oil (0.002 g, 6%). TLC R_f 0.2 (10% MeOH/DCM). LC−MS (ESI) *m*/*z*: 360 $[M + H]^+$. ^{1}H NMR (400 MHz, MeOD) δ 10.18 (t, *J* = 1.7 Hz, 1H), 10.00 (dt, *J* = 6.3, 1.4 Hz, 1H), 9.05 (dt, *J* = 8.1, 1.6 Hz, 1H), 8.31 (dd, *J* = 8.1, 6.3 Hz, 1H), 8.25 (d, *J* = 6.0 Hz, 1H), 7.38 (t, *J* = 7.9 Hz, 1H), 7.03−7.08 (m, 1H), 7.02 (t, *J* = 2.1 Hz, 1H), 6.89−6.79 (m, 1H), 6.73 (d, *J* = 6.0 Hz, 1H), 4.22−3.97 (m, 1H), 2.89 (s, 3H), 2.11 (d, *J* = 13.3 Hz, 2H), 1.87 (dt, *J* = 13.0, 3.7 Hz, 2H), 1.74 (dt, *J* = 12.9, 3.8 Hz, 1H), 1.58−1.27 (m, 5H). ^{13}C NMR (101 MHz, MeOD) δ 163.12, 149.09, 148.41, 139.93, 137.95, 136.60, 131.55, 127.82, 127.13, 126.40, 125.55, 76.82, 51.05, 39.06, 33.48, 26.68, 26.09.

*N^4-Cyclohexyl-N^2,N^4-dimethyl-N^2-(3-(pyridin-3-yl)phenyl)-pyrimidine-2,4-diamine (**17**).* To a mixture of **15** (0.10 mmol, 1.0 equiv) in EtOH (0.50 mL, 0.20 M) under a nitrogen atmosphere at rt were added *N*-methyl-3-(pyridin-3-yl)aniline (0.11 mmol, 1.1 equiv) and a drop of 1 N HCl. The reaction mixture was heated to reflux and stirred at that temperature for 3 h; it was then cooled to rt. Purification using automated flash chromatography (MeOH/DCM) was followed by evaporation, giving **17** as an orange oil (0.010 g, 27%). TLC R_f 0.4 (10% MeOH/DCM). LC−MS (ESI) *m*/*z*: 374 $[M + H]^+$. ^{1}H NMR (500 MHz, MeOD) δ 10.19 (s, 1H), 10.01 (d, *J* = 7.4 Hz, 1H), 9.06 (d, *J* = 8.1 Hz, 1H), 8.42 (t, *J* = 7.2 Hz, 1H), 8.33 (t, *J* = 7.2 Hz, 1H), 7.38 (t, *J* = 7.9 Hz, 1H), 7.08 (d, *J* = 7.5 Hz, 1H), 7.03 (s,

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J = 2.0 Hz, 1H), 6.98−6.88 (m, 1H), 6.84 (d, J = 5.0 Hz, 1H), 3.95 (s, 1H), 3.09−3.26 (m, 3H), 2.89 (s, 3H), 2.02−1.89 (m, 2H), 1.83−1.86 (m, 2H), 1.67−1.77 (m, 3H), 1.56 (qt, J = 13.2, 3.6 Hz, 2H), 1.38−1.23 (m, 1H). ^{13}C NMR (126 MHz, MeOD) δ 162.46, 156.01, 155.64, 151.30, 146.51, 141.80, 138.18, 137.88, 134.24, 130.11, 127.39, 114.72, 114.23, 109.54, 105.17, 55.20, 29.23, 29.05, 25.53, 25.14.

*2-Chloro-4-(cyclohexyloxy)pyrimidine (**18**).* To a solution of cyclohexanol (6.0 mmol, 1.2 equiv) in DMF (10 mL, 0.60 M) at 0 °C was added NaH as a 60% suspension in mineral oil (7.5 mmol, 1.5 equiv). After stirring for 20 min, 2,4-dichloropyrimidine **2** (5.0 mmol, 1.0 equiv) was added at 0 °C to one portion. The reaction mixture was then stirred under a nitrogen atmosphere while warming to rt over 16 h. Brine (20 mL) was added to the reaction mixture; then, the mixture was extracted into EtOAc (2 × 20 mL). The organic phase was dried over magnesium sulfate, filtered, and concentrated. Purification using automated flash chromatography (EtOAc/hexanes) was followed by evaporation, giving **18** as a white solid (0.031 g, 3%). TLC R_f 0.35 (10% EtOAc/hexanes). LC−MS (ESI) m/z: 213 $[M + H]^+$. ^{1}H NMR (400 MHz, $CDCl_3$) δ 8.18 (d, J = 5.7 Hz, 1H), 6.53 (d, J = 5.7 Hz, 1H), 5.17−4.97 (m, 1H), 2.00−1.84 (m, 2H), 1.78−1.63 (m, 2H), 1.56−1.49 (m, 2H), 1.35−1.48 (m, 3H), 1.19−1.34 (m, 1H). ^{13}C NMR (101 MHz, $CDCl_3$) δ 170.02, 160.21, 158.63, 107.61, 75.52, 31.32, 25.36, 23.60.

*4-(Cyclohexyloxy)-N-(3-(pyridin-3-yl)phenyl)pyrimidin-2-amine (**19**).* To a mixture of **18** (0.10 mmol, 1.0 equiv) in EtOH (0.50 mL, 0.20 M) under a nitrogen atmosphere at rt were added 3-(pyridin-3-yl)aniline (0.11 mmol, 1.1 equiv) and a drop of 1 N HCl. The reaction mixture was heated to reflux and stirred at that temperature for 3 h; it was then cooled to rt. Purification using automated flash chromatography (MeOH/DCM) was followed by evaporation, giving **19** as an orange oil (0.003 g, 9%). TLC R_f 0.3 (10% MeOH/DCM). LC−MS (ESI) m/z: 347 $[M + H]^+$. ^{1}H NMR (500 MHz, MeOD) δ 10.17 (d, J = 1.8 Hz, 1H), 10.02 (dt, J = 6.3, 1.8 Hz, 1H), 9.09 (dt, J = 8.1, 2.2 Hz, 1H), 8.79 (t, J = 6.1 Hz, 1H), 8.37 (dd, J = 8.0, 6.3 Hz, 1H), 7.36 (t, J = 7.8 Hz, 1H), 7.27−7.07 (m, 3H), 6.94 (dd, J = 8.0, 2.3 Hz, 1H), 5.40−5.45 (m, 1H), 2.13−2.18 (m, 2H), 1.96−1.83 (m, 2H), 1.70−1.78 (m, 3H), 1.65−1.53 (m, 2H), 1.44−1.49 (m, 1H). ^{13}C NMR (126 MHz, MeOD) δ 171.00, 159.02, 154.69, 149.51, 147.06, 141.78, 138.57, 138.14, 134.01, 130.30, 127.67, 116.71, 115.78, 112.77, 111.06, 77.04, 30.86, 25.03, 23.13.

*N-Cyclohexyl-2-(3-(pyridin-3-yl)phenoxy)pyrimidin-4-amine (**22**).* To a mixture of **3a** (0.10 mmol, 1.0 equiv) in EtOH (0.50 mL, 0.20 M) under a nitrogen atmosphere at rt were added 3-(pyridin-3-yl)phenol (0.11 mmol, 1.1 equiv) and a drop of 1 N HCl. The reaction mixture was heated to reflux and stirred at that temperature for 16 h; it was then cooled to rt. Purification using automated flash chromatography (MeOH/DCM) was followed by evaporation, giving **22** as a yellow oil (0.028 g, 81%). TLC R_f 0.1 (10% MeOH/DCM). LC−MS (ESI) m/z: 347 $[M + H]^+$. ^{1}H NMR (500 MHz, MeOD) δ 10.19 (d, J = 9.9 Hz, 1H), 10.01 (d, J = 10.0 Hz, 1H), 9.05 (d, J = 9.1 Hz, 1H), 8.29 (dt, J = 41.0, 6.6 Hz, 2H), 7.46 (t, J = 9.8 Hz, 1H), 7.40−7.23 (m, 2H), 7.04 (d, J = 9.0 Hz, 1H), 6.82−6.65 (m, 1H), 4.10 (m, 1H), 2.11 (m, 2H), 2.00−1.81 (m, 2H), 1.73 (m, 1H), 1.64−1.26 (m, 5H). ^{13}C NMR (126 MHz, MeOD) δ 162.96, 158.68, 154.25, 146.51, 140.85, 138.31, 137.88, 134.78, 130.67, 127.49, 117.99, 117.09, 113.84, 107.95, 49.88, 31.93, 25.33, 24.58.

*4-(Cyclohexyloxy)-2-(3-(pyridin-3-yl)phenoxy)pyrimidine (**20**).* To a mixture of **18** (0.10 mmol, 1.0 equiv) in EtOH (0.50 mL, 0.20 M) under a nitrogen atmosphere at rt were added 3-(pyridin-3-yl)phenol (0.11 mmol, 1.1 equiv) and a drop of 1 N HCl. The reaction mixture was heated to reflux and stirred at that temperature for 16 h; it was then cooled to rt. Purification using automated flash chromatography (MeOH/DCM) was followed by evaporation, giving **20** as a yellow solid (0.018 g, 52%). TLC R_f 0.3 (10% MeOH/DCM). LC−MS (ESI) m/z: 348 $[M + H]^+$. ^{1}H NMR (400 MHz, MeOD) δ 10.19 (t, J = 1.7 Hz, 1H), 10.05 (dt, J = 6.4, 1.4 Hz, 1H), 9.13 (ddd, J = 8.1, 2.0, 1.2 Hz, 1H), 8.79 (d, J = 5.8 Hz, 1H), 8.39 (dd, J = 8.1, 6.4 Hz, 1H), 7.48 (t, J = 7.9 Hz, 1H), 7.36 (ddd, J = 7.7, 1.9, 1.0 Hz, 1H), 7.31 (t, J = 2.1 Hz, 1H), 7.21 (d, J = 5.8 Hz, 1H), 7.06 (ddd, J = 8.2, 2.4, 0.9 Hz, 1H), 5.42−5.45 (m, 1H), 2.25−2.07 (m, 2H), 1.86−1.92 (m, 2H), 1.67−1.78 (m, 3H), 1.64−1.52 (m, 2H), 1.44−1.49 (m, 1H). ^{13}C NMR (101 MHz, MeOD) δ 172.42, 160.45, 160.12, 148.65, 142.60, 140.21, 139.72, 136.02, 132.14, 129.17, 119.49, 118.62, 115.32, 112.51, 78.48, 32.28, 26.45, 24.56.

*N^4-Cyclohexyl-N^2-phenylpyrimidine-2,4-diamine (**4a**).* To a mixture of **3a** (0.10 mmol, 1.0 equiv) in EtOH (0.50 mL, 0.20 M) under a nitrogen atmosphere at rt were added aniline (0.20 mmol, 2.0 equiv) and a drop of 1 N HCl. The reaction mixture was heated to reflux and stirred at that temperature for 1.5 h; it was then cooled to rt. Purification using automated flash chromatography (MeOH/DCM) was followed by evaporation, giving **4a** as a white solid (0.018 g, 67%). TLC R_f 0.7 (10% MeOH/DCM). LC−MS (ESI) m/z: 269 $[M + H]^+$. ^{1}H NMR (500 MHz, $CDCl_3$) δ 7.90−7.72 (m, 1H), 7.53 (d, J = 7.9 Hz, 2H), 7.25−7.20 (m, 2H), 7.19 (br s, 1H), 6.92 (t, J = 7.4 Hz, 1H), 5.75 (d, J = 5.9 Hz, 1H), 4.71 (br s, 1H), 3.60 (br s, 1H), 2.05−1.92 (m, 2H), 1.69−1.75 (m, 2H), 1.58−1.62 (m, 1H), 1.29−1.38 (m, 2H), 1.21−1.08 (m, 3H). ^{13}C NMR (126 MHz, $CDCl_3$) δ 162.03, 159.44, 139.97, 128.73, 121.97, 119.23, 50.07, 33.09, 25.62, 24.93.

*N^2-([1,1′-Biphenyl]-2-yl)-N^4-cyclohexylpyrimidine-2,4-diamine (**4b**).* To a mixture of **3a** (0.10 mmol, 1.0 equiv) in EtOH (0.50 mL, 0.20 M) under a nitrogen atmosphere at rt were added 2-aminobiphenyl (0.20 mmol, 2.0 equiv) and a drop of 1 N HCl. The reaction mixture was heated to reflux and stirred at that temperature for 1.5 h; it was then cooled to rt. Purification using automated flash chromatography (MeOH/DCM) was followed by evaporation, giving **4b** as a colorless oil (0.023 g, 67%). TLC R_f 0.45 (5% MeOH/DCM). LC−MS (ESI) m/z: 345 $[M + H]^+$. ^{1}H NMR (400 MHz, $CDCl_3$) δ 8.35 (d, J = 8.3 Hz, 1H), 7.78 (s, 1H), 7.51−7.39 (m, 4H), 7.32−7.37 (m, 2H), 7.29−7.20 (m, 2H), 7.09 (td, J = 7.5, 1.2 Hz, 1H), 5.81 (d, J = 6.0 Hz, 1H), 4.97 (br s, 1H), 3.60 (br s, 1H), 1.98−2.04 (m, 2H), 1.74−1.80 (m, 2H), 1.63−1.68 (m, 1H), 1.44−1.29 (m, 2H), 1.15−1.27 (m, 3H). ^{13}C NMR (101 MHz, $CDCl_3$) δ 161.95, 138.81, 136.42, 132.54, 130.24, 129.40, 128.92, 127.85, 127.59, 122.62, 121.18, 50.14, 33.00, 25.56, 24.87.

*N^4-Cyclohexyl-N^2-(3-cyclopropylphenyl)pyrimidine-2,4-diamine (**4c**).* To a mixture of **3a** (0.10 mmol, 1.0 equiv) in EtOH (0.50 mL, 0.20 M) under a nitrogen atmosphere at rt were added 3-cyclopropylaniline (0.11 mmol, 1.1 equiv) and a drop of 1 N HCl. The reaction mixture was heated to reflux and stirred at that temperature for 4 h; it was then cooled to rt. Purification using automated flash chromatography (MeOH/DCM) was followed by evaporation, giving **4c** as a white oil (0.009 g, 29%). TLC R_f 0.6 (10% MeOH/DCM). LC−MS (ESI) m/z: 309 $[M + H]^+$. ^{1}H NMR (500 MHz, $CDCl_3$) δ 7.76 (d, J = 6.2 Hz, 1H), 7.35−7.37 (m, 2H), 7.20 (s, 1H), 7.10 (t, J = 7.9 Hz, 1H), 6.63 (d, J = 5.0 Hz, 1H), 5.76 (d, J = 6.0 Hz, 1H), 4.88 (d, J = 8.0 Hz, 1H), 3.77−3.46 (m, 1H), 1.95−2.00 (m, 2H), 1.80−1.83 (m, 1H), 1.70 (dt, J = 13.6, 3.9 Hz, 2H), 1.59 (dt, J = 13.0, 3.8 Hz, 1H), 1.40−1.24 (m,

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2H), 1.24−1.09 (m, 3H), 0.91−0.79 (m, 2H), 0.70−0.56 (m, 2H). ^{13}C NMR (126 MHz, $CDCl_3$) δ 162.02, 158.97, 153.51, 144.68, 139.65, 128.60, 119.35, 117.17, 116.73, 96.62, 49.98, 33.06, 25.58, 24.88, 15.58, 9.15.

*N^4-Cyclohexyl-N^2-(3-fluorophenyl)pyrimidine-2,4-diamine (**4d**).* To a mixture of **3a** (0.10 mmol, 1.0 equiv) in EtOH (0.50 mL, 0.20 M) at rt were added 3-fluoroaniline (0.20 mmol, 2.0 equiv) and a drop of 1 N HCl. The reaction mixture was heated to reflux and stirred at that temperature for 1.5 h; it was then cooled to rt. Purification using automated flash chromatography (MeOH/DCM) was followed by evaporation, giving **4d** as a white solid (0.018 g, 63%). TLC R_f 0.7 (10% MeOH/DCM). LC−MS (ESI) m/z: 287 $[M + H]^+$. ^{1}H NMR (500 MHz, $CDCl_3$) δ 7.90 (d, J = 6.0 Hz, 1H), 7.78 (d, J = 12.0 Hz, 1H), 7.30 (s, 1H), 7.19 (dd, J = 8.2, 6.7 Hz, 1H), 7.07 (dd, J = 8.1, 2.1 Hz, 1H), 6.66 (td, J = 8.3, 2.5 Hz, 1H), 5.85 (d, J = 5.8 Hz, 1H), 4.76 (br s, 1H), 3.72 (br s, 1H), 2.16−2.00 (m, 2H), 1.77−1.81 (m, 2H), 1.66−1.70 (m, 1H), 1.50−1.35 (m, 2H), 1.19−1.27 (m, 3H). ^{13}C NMR (126 MHz, $CDCl_3$) δ 164.17, 162.25, 161.98, 159.34, 141.88, 141.79, 129.65, 129.57, 114.11, 114.08, 108.21, 108.04, 106.15, 105.93, 50.12, 33.11, 25.62, 24.90.

*N^4-Cyclohexyl-N^2-(pyridin-3-yl)pyrimidine-2,4-diamine (**4e**).* To a mixture of **3a** (0.10 mmol, 1.0 equiv) in EtOH (0.50 mL, 0.20 M) under a nitrogen atmosphere at rt were added 3-aminopyridine (0.20 mmol, 2.0 equiv) and a drop of 1 N HCl. The reaction mixture was heated to reflux and stirred at that temperature for 1.5 h; it was then cooled to rt. Purification using automated flash chromatography (MeOH/DCM) was followed by evaporation, giving **4e** as a yellow oil (0.023 g, 85%). TLC R_f 0.4 (10% MeOH/DCM). LC−MS (ESI) m/z: 270 $[M + H]^+$. ^{1}H NMR (400 MHz, MeOD) δ 9.29 (d, J = 2.3 Hz, 1H), 9.17 (dt, J = 5.1, 1.7 Hz, 1H), 8.17 (d, J = 6.0 Hz, 1H), 7.95−7.76 (m, 2H), 6.67 (d, J = 6.0 Hz, 1H), 4.22−3.98 (m, 1H), 2.11−1.96 (m, 2H), 1.82−1.87 (m, 2H), 1.78−1.66 (m, 1H), 1.48−1.55 (m, 2H), 1.44−1.19 (m, 3H). ^{13}C NMR (126 MHz, $CDCl_3$) δ 166.85, 159.11, 158.07, 152.94, 134.51, 131.01, 128.10, 111.57, 53.28, 35.98, 29.22, 28.41.

*Methyl 3-((4-(cyclohexylamino)pyrimidin-2-yl)amino)-benzoate (**4f**).* To a mixture of **3a** (0.10 mmol, 1.0 equiv) in MeOH (0.50 mL, 0.20 M) under a nitrogen atmosphere at rt were added 3-aminobenzoic acid methyl ester (0.11 mmol, 1.1 equiv) and a drop of 1 N HCl. The reaction mixture was heated to reflux and stirred at that temperature for 2 h; it was then cooled to rt. Purification using automated flash chromatography (MeOH/DCM) was followed by evaporation, giving **4f** as a colorless oil (0.018 g, 55%). TLC R_f 0.6 (10% MeOH/DCM). LC−MS (ESI) m/z: 327 $[M + H]^+$. ^{1}H NMR (500 MHz, $CDCl_3$) δ 8.21 (s, 1H), 7.98−7.82 (m, 2H), 7.66 (d, J = 5.0 Hz, 1H), 7.61 (s, 1H), 7.35 (t, J = 7.9 Hz, 1H), 5.86 (d, J = 5.9 Hz, 1H), 4.88 (br s, 1H), 3.91 (s, 3H), 3.08 (br s, 1H), 2.11−1.96 (m, 2H), 1.76 (dt, J = 13.7, 3.9 Hz, 2H), 1.70−1.60 (m, 1H), 1.47−1.31 (m, 2H), 1.19−1.25 (m, 3H). ^{13}C NMR (126 MHz, $CDCl_3$) δ 167.13, 162.02, 159.21, 154.87, 140.20, 130.65, 128.74, 123.60, 123.02, 120.18, 52.12, 49.94, 33.05, 25.60, 24.80.

*N^2-([1,1′-Biphenyl]-3-yl)-N^4-cyclohexylpyrimidine-2,4-diamine (**4g**).* To a mixture of **3a** (0.10 mmol, 1.0 equiv) in EtOH (0.50 mL, 0.20 M) under a nitrogen atmosphere at rt were added 3-aminobiphenyl (0.20 mmol, 2.0 equiv) and a drop of 1 N HCl. The reaction mixture was heated to reflux and stirred at that temperature for 1.5 h; it was then cooled to rt. Purification using automated flash chromatography (MeOH/DCM) was followed by evaporation, giving **4g** as a white solid (0.018 g, 52%). TLC R_f 0.5 (5% MeOH/DCM). LC−MS (ESI) m/z: 345 $[M + H]^+$. ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.84 (s, 2H), 7.59−7.62 (m, 3H), 7.43 (dd, J = 8.2, 6.9 Hz, 2H), 7.37−7.31 (m, 2H), 7.23 (dt, J = 7.7, 1.4 Hz, 1H), 5.86 (d, J = 6.0 Hz, 1H), 4.97 (d, J = 8.0 Hz, 1H), 3.71 (s, 1H), 2.10−1.92 (m, 2H), 1.77−1.64 (m, 2H), 1.59 (d, J = 11.9 Hz, 1H), 1.36−1.09 (m, 5H). ^{13}C NMR (101 MHz, $CDCl_3$) δ 162.02, 142.00, 141.39, 140.04, 129.07, 128.67, 127.28, 121.29, 118.42, 50.06, 33.02, 25.53, 24.73.

*N^4-Cyclohexyl-N^2-(3-(pyridin-2-yl)phenyl)pyrimidine-2,4-diamine (**4h**).* To a mixture of **3a** (0.10 mmol, 1.0 equiv) in EtOH (0.50 mL, 0.20 M) under a nitrogen atmosphere at rt were added 3-(pyridin-2-yl)aniline (0.11 mmol, 1.1 equiv) and a drop of 1 N HCl. The reaction mixture was heated to reflux and stirred at that temperature for 3 h; it was then cooled to rt. Purification using automated flash chromatography (MeOH/DCM) was followed by evaporation, giving **4h** as a white film (0.007 g, 20%). TLC R_f 0.5 (10% MeOH/DCM). LC−MS (ESI) m/z: 346 $[M + H]^+$. ^{1}H NMR (500 MHz, MeOD) δ 8.74−8.58 (m, 1H), 8.27 (s, 1H), 7.95 (td, J = 7.7, 1.8 Hz, 1H), 7.89−7.94 (m, 1H), 7.73 (d, J = 7.4 Hz, 1H), 7.70−7.63 (m, 1H), 7.60−7.55 (m, 1H), 7.52 (t, J = 7.8 Hz, 1H), 7.41−7.43 (m, 1H), 6.14 (d, J = 7.0 Hz, 1H), 3.90 (s, 1H), 2.06−1.89 (m, 2H), 1.74−1.60 (m, 2H), 1.60−1.49 (m, 1H), 1.30−1.21 (m, 2H), 1.21−1.05 (m, 3H). ^{13}C NMR (126 MHz, MeOD) δ 161.97, 157.24, 154.20, 149.00, 143.49, 140.02, 138.14, 137.66, 122.94, 122.63, 122.01, 121.48, 120.39, 98.62, 50.18, 31.86, 25.07, 24.52.

*N^4-Cyclohexyl-N^2-(3′-methoxy-[1,1′-biphenyl]-3-yl)-pyrimidine-2,4-diamine (**4i**).* To a mixture of **3a** (0.10 mmol, 1.0 equiv) in EtOH (0.50 mL, 0.20 M) under a nitrogen atmosphere at rt were added (3′-methoxybiphenyl-3-yl)amine HCl (0.11 mmol, 1.1 equiv) and a drop of 1 N HCl. The reaction mixture was heated to reflux and stirred at that temperature for 3 h; it was then cooled to rt. Purification using automated flash chromatography (EtOAc/hexanes) was followed by evaporation, giving **4i** as a colorless oil (0.007 g, 19%). TLC R_f 0.2 (50% EtOAc/hexanes + 1% MeOH). LC−MS (ESI) m/z: 375 $[M + H]^+$. ^{1}H NMR (500 MHz, $CDCl_3$) δ 7.98−7.89 (m, 1H), 7.85 (s, 1H), 7.63−7.65 (m, 1H), 7.35−7.37 (m, 2H), 7.21−7.24 (m, 2H), 7.18 (s, 1H), 7.15−7.16 (m, 1H), 6.90−6.93 (m, 1H), 5.85 (d, J = 5.9 Hz, 1H), 4.71 (s, 1H), 3.89 (s, 3H), 3.84−3.57 (m, 1H), 2.03−2.07 (m, 2H), 1.81−1.68 (m, 2H), 1.68−1.56 (m, 1H), 1.39−1.27 (m, 2H), 1.17−1.25 (m, 3H). ^{13}C NMR (126 MHz, $CDCl_3$) δ 162.06, 159.85, 159.82, 155.82, 143.05, 141.85, 140.49, 129.64, 129.03, 120.89, 119.83, 118.28, 118.07, 112.97, 112.67, 95.81, 55.30, 49.85, 33.12, 25.61, 24.74.

*Methyl 3′-((4-(cyclohexylamino)pyrimidin-2-yl)amino)-[1,1′-biphenyl]-3-carboxylate (**4j**).* To a mixture of **3a** (0.10 mmol, 1.0 equiv) in EtOH (0.50 mL, 0.20 M) under a nitrogen atmosphere at rt were added 3′-aminobiphenyl-3-carboxylic acid methyl ester HCl (0.11 mmol, 1.1 equiv) and a drop of 1 N HCl. The reaction mixture was heated to reflux and stirred at that temperature for 3 h; it was then cooled to rt. Purification using automated flash chromatography (MeOH/DCM) was followed by evaporation, giving **4j** as a white solid (0.012 g, 30%). TLC R_f 0.2 (50% EtOAc/hexanes + 1% MeOH). LC−MS (ESI) m/z: 375 $[M + H]^+$. ^{1}H NMR (500 MHz, $CDCl_3$) δ 8.31 (t, J = 1.8 Hz, 1H), 8.04 (dt, J = 7.8, 1.4 Hz, 1H), 7.97−7.87 (m, 2H), 7.82 (ddd, J = 7.8, 1.9, 1.2 Hz, 1H), 7.64 (ddd, J = 8.1, 2.3, 1.0 Hz, 1H), 7.53 (t, J = 7.7 Hz, 1H), 7.40 (t, J = 7.9 Hz, 1H), 7.26 (ddd, J = 7.7, 1.8, 1.0 Hz, 1H), 7.20 (s, 1H), 5.86 (d, J = 5.9 Hz, 1H), 4.74 (br s, 1H), 3.97 (br s, 3H), 3.86−3.56 (m, 1H), 2.11−1.99 (m, 2H), 1.78−1.67 (m, 2H), 1.61 (d, J = 11.2 Hz, 1H), 1.36−1.13 (m, 5H). ^{13}C NMR (126 MHz, $CDCl_3$) δ 167.09, 162.04, 159.62, 155.39, 141.74, 140.83, 140.59, 131.68, 130.59, 129.22, 128.77,

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128.42, 128.31, 120.85, 118.56, 117.98, 77.27, 52.19, 49.89, 33.08, 25.56, 24.75.

*N^4-Cyclohexyl-N^2-(3-(oxazol-2-yl)phenyl)pyrimidine-2,4-diamine (**4k**).* To a mixture of **3a** (0.10 mmol, 1.0 equiv) in EtOH (0.50 mL, 0.20 M) under a nitrogen atmosphere at rt were added 3-(1,3-oxaxol-2-yl)aniline (0.11 mmol, 1.1 equiv) and a drop of 1 N HCl. The reaction mixture was heated to reflux and stirred at that temperature for 2 h; it was then cooled to rt. Purification using automated flash chromatography (MeOH/DCM) was followed by evaporation, giving **4k** as a white solid (0.022 g, 66%). TLC R_f 0.6 (10% MeOH/DCM). LC−MS (ESI) m/z: 336 $[M + H]^+$. ^{1}H NMR (500 MHz, $CDCl_3$) δ 8.40 (s, 1H), 7.97−7.83 (m, 1H), 7.76−7.65 (m, 3H), 7.41 (s, 1H), 7.28 (s, 1H), 7.25 (s, 1H), 5.89 (d, J = 6.0 Hz, 1H), 5.04 (br s, 1H), 3.74 (br s, 1H), 2.13−2.01 (m, 2H), 1.76−1.80 (m, 2H), 1.65−1.69 (m, 1H), 1.47−1.34 (m, 2H), 1.21−1.29 (m, 3H). ^{13}C NMR (126 MHz, $CDCl_3$) δ 162.08, 162.02, 158.70, 140.30, 138.48, 129.23, 128.37, 127.92, 121.20, 120.13, 117.01, 50.03, 33.08, 25.57, 24.81.

*N^2-(3-(1H-Pyrazol-3-yl)phenyl)-N^4-cyclohexylpyrimidine-2,4-diamine (**4l**).* To a mixture of **3a** (0.10 mmol, 1.0 equiv) in EtOH (0.50 mL, 0.20 M) under a nitrogen atmosphere at rt were added 3-(1*H*-pyrazol-3-yl)aniline (0.11 mmol, 1.1 equiv) and a drop of 1 N HCl. The reaction mixture was heated to reflux and stirred at that temperature for 2 h; it was then cooled to rt. Purification using automated flash chromatography (MeOH/DCM) was followed by evaporation, giving **4l** as a colorless oil (0.020 g, 60%). TLC R_f 0.6 (10% MeOH/DCM). LC−MS (ESI) m/z: 335 $[M + H]^+$. ^{1}H NMR (500 MHz, MeOD) δ 8.08 (s, 1H), 7.69 (d, J = 16.6 Hz, 2H), 7.56 (d, J = 8.0 Hz, 1H), 7.43 (d, J = 8.0 Hz, 1H), 7.36 (td, J = 7.8, 1.8 Hz, 1H), 6.67 (d, J = 2.5 Hz, 1H), 5.99 (dd, J = 6.4, 1.8 Hz, 1H), 3.92 (s, 1H), 2.00−2.02 (m, 2H), 1.72−1.75 (m, 2H), 1.61−1.63 (m, 1H), 1.19−1.31 (m, 5H). ^{13}C NMR (126 MHz, MeOD) δ 162.23, 158.06, 150.32, 140.11, 128.66, 119.78, 119.48, 117.56, 102.17, 97.88, 49.48, 32.36, 25.35, 24.70.

*N^2-(3-(1H-Imidazol-1-yl)phenyl)-N^4-cyclohexylpyrimidine-2,4-diamine (**4m**).* To a mixture of **3a** (0.10 mmol, 1.0 equiv) in EtOH (0.50 mL, 0.20 M) under a nitrogen atmosphere at rt were added 3-(1*H*-imidazol-1-yl)aniline (0.11 mmol, 1.1 equiv) and a drop of 1 N HCl. The reaction mixture was heated to reflux and stirred at that temperature for 20 h; it was then cooled to rt. Purification using automated reversed-phase flash chromatography (water/MeOH) was followed by evaporation, giving **4m** as a colorless oil (0.010 g, 30%). LC−MS (ESI) m/z: 335 $[M + H]^+$. ^{1}H NMR (400 MHz, MeOD) δ 8.50 (br s, 1H), 8.46 (d, J = 2.2 Hz, 1H), 8.16−8.01 (m, 2H), 7.35 (t, J = 8.0 Hz, 1H), 7.06−6.95 (m, 2H), 6.91 (ddd, J = 8.2, 2.1, 0.9 Hz, 1H), 6.56 (d, J = 6.1 Hz, 1H), 4.13−4.14 (m, 1H), 2.11−1.97 (m, 2H), 1.81−1.86 (m, 2H), 1.70−1.74 (m, 1H), 1.57−1.44 (m, 2H), 1.36−1.39 (m, 3H). ^{13}C NMR (101 MHz, MeOD) δ 164.23, 155.53, 153.70, 151.78, 137.14, 131.98, 123.60, 120.79, 117.54, 111.20, 108.63, 107.95, 50.50, 33.57, 26.75, 25.94.

*N^4-Cyclohexyl-N^2-(3-(pyrazin-2-yl)phenyl)pyrimidine-2,4-diamine (**4n**).* To a mixture of **3a** (0.10 mmol, 1.0 equiv) in EtOH (0.50 mL, 0.20 M) under a nitrogen atmosphere at rt were added 3-(pyrazin-2-yl)aniline (0.11 mmol, 1.1 equiv) and a drop of 1 N HCl. The reaction mixture was heated to reflux and stirred at that temperature for 5 h; it was then cooled to rt. Purification using automated flash chromatography (MeOH/EtOAc) was followed by evaporation, giving **4n** as a colorless oil (0.007 g, 20%). TLC R_f 0.7 (10% MeOH/EtOAc). LC−MS (ESI) m/z: 347 $[M + H]^+$. ^{1}H NMR (400 MHz, MeOD) δ 9.13 (d, J = 1.6 Hz, 1H), 8.75−8.67 (m, 1H), 8.57 (d, J = 2.5 Hz, 1H), 8.43 (s, 1H), 7.72 (t, J = 8.7 Hz, 3H), 7.49 (t, J = 7.9 Hz, 1H), 6.02 (d, J = 6.3 Hz, 1H), 3.91 (s, 1H), 1.99−2.03 (m, 2H), 1.70−1.72 (m, 2H), 1.60 (s, 1H), 1.37−1.13 (m, 5H). ^{13}C NMR (101 MHz, MeOD) δ 163.62, 158.67, 154.55, 145.64, 144.08, 143.40, 141.62, 138.07, 130.47, 123.02, 122.50, 120.22, 101.40, 51.02, 33.61, 26.67, 26.06.

*N^4-Cyclohexyl-N^2-(3-morpholinophenyl)pyrimidine-2,4-diamine (**4o**).* To a mixture of **3a** (0.10 mmol, 1.0 equiv) in EtOH (0.50 mL, 0.20 M) under a nitrogen atmosphere at rt were added 3-morpholin-4-ylaniline (0.11 mmol, 1.1 equiv) and a drop of 1 N HCl. The reaction mixture was heated to reflux and stirred at that temperature for 3 h; it was then cooled to rt. Purification using automated flash chromatography (MeOH/DCM) was followed by evaporation, giving **4o** as a white oil (0.018 g, 51%). TLC R_f 0.4 (5% MeOH/DCM). LC−MS (ESI) m/z: 354 $[M + H]^+$. ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.62 (s, 1H), 7.21 (d, J = 6.6 Hz, 3H), 7.11−7.02 (m, 1H), 6.64 (dd, J = 5.7, 3.4 Hz, 1H), 5.98 (d, J = 6.7 Hz, 1H), 3.91−3.70 (m, 5H), 3.22−3.10 (m, 4H), 2.10−1.93 (m, 2H), 1.76−1.82 (m, 2H), 1.64−1.69 (m, 1H), 1.46−1.12 (m, 5H). ^{13}C NMR (101 MHz, $CDCl_3$) δ 161.86, 151.96, 138.95, 129.33, 112.24, 111.16, 107.93, 66.89, 50.64, 49.30, 32.63, 25.36, 24.77.

*N^4-Cyclohexyl-N^2-(1-phenylpiperidin-3-yl)pyrimidine-2,4-diamine (**4p**).* To a mixture of **3a** (0.10 mmol, 1.0 equiv) in EtOH (0.50 mL, 0.20 M) under a nitrogen atmosphere at rt were added 1-phenylpiperidin-3-amine (0.11 mmol, 1.1 equiv) and a drop of 1 N HCl. The reaction mixture was heated to reflux and stirred at that temperature for 22 h; it was then cooled to rt. Purification using automated flash chromatography (MeOH/EtOAc) was followed by evaporation, giving **4p** as a colorless oil (0.002 g, 6%). TLC R_f 0.3 (10% MeOH/EtOAc). LC−MS (ESI) m/z: 352 $[M + H]^+$. ^{1}H NMR (500 MHz, MeOD) δ 7.58 (s, 1H), 7.23 (t, J = 10.0 Hz, 2H), 7.00 (d, J = 8.1 Hz, 2H), 6.83 (t, J = 7.3 Hz, 1H), 5.84 (d, J = 6.5 Hz, 1H), 4.17−4.03 (m, 1H), 3.87 (s, 1H), 3.70 (d, J = 11.6 Hz, 1H), 3.44−3.36 (m, 1H), 2.93 (t, J = 15.0 Hz, 1H), 2.82−2.73 (m, 1H), 1.98 (d, J = 8.1 Hz, 3H), 1.96−1.86 (m, 1H), 1.79−1.71 (m, 2H), 1.58−1.65 (m, 2H), 1.31−1.40 (m, 2H), 1.20−1.29 (m, 3H). ^{13}C NMR (126 MHz, MeOD) δ 162.32, 151.98, 128.59, 119.61, 116.99, 55.38, 50.15, 49.16, 32.45, 32.40, 29.61, 25.36, 24.67, 23.24.

*N^4-Cyclohexyl-N^2-(3-(pyridin-3-yl)cyclohexyl)pyrimidine-2,4-diamine (**4q**).* To a mixture of **3a** (0.080 mmol, 1.0 equiv) in EtOH (0.50 mL, 0.16M) under a nitrogen atmosphere at rt were added 3-(pyridin-3-yl)cyclohexan-1-amine (0.080 mmol, 1.0 equiv) and a drop of 1 N HCl. The reaction mixture was heated to reflux and stirred at that temperature for 6 h; it was then cooled to rt. Purification using automated flash chromatography (MeOH/DCM) was followed by evaporation, giving **4q** as a yellow oil (0.002 g, 7%). TLC R_f 0.2 (10% MeOH/DCM). LC−MS (ESI) m/z: 353 $[M + H]^+$. ^{1}H NMR (500 MHz, MeOD) δ 9.91 (d, J = 7.0 Hz, 2H), 8.79 (d, J = 8.0 Hz, 1H), 8.22 (q, J = 5.0 Hz, 2H), 6.70 (d, J = 6.0 Hz, 1H), 4.18−4.04 (m, 1H), 3.78 (td, J = 10.8, 5.4 Hz, 1H), 3.11 (t, J = 12.3 Hz, 1H), 2.28 (d, J = 11.3 Hz, 1H), 2.13−2.05 (m, 3H), 2.05−1.96 (m, 2H), 1.84−1.89 (m, 2H), 1.74 (dt, J = 12.8, 4.1 Hz, 1H), 1.48−1.63 (m, 4H), 1.44−1.28 (m, 4H). ^{13}C NMR (126 MHz, MeOD) δ 162.95, 155.05, 154.22, 147.78, 146.83, 138.49, 138.24, 127.22, 107.83, 69.34, 49.71, 41.40, 39.97, 34.25, 32.11, 31.96, 25.32, 24.56, 23.72.

*N^4-Cyclohexyl-N^2-(4-fluorophenyl)pyrimidine-2,4-diamine (**4r**).* To a mixture of **3a** (0.10 mmol, 1.0 equiv) in EtOH (0.50 mL, 0.20 M) under a nitrogen atmosphere at rt were added 4-fluoroaniline (0.20 mmol, 2.0 equiv) and a drop of 1 N HCl. The

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reaction mixture was heated to reflux and stirred at that temperature for 1.5 h; it was then cooled to rt. Purification using automated flash chromatography (MeOH/DCM) was followed by evaporation, giving **4r** as a white solid (0.024 g, 84%). TLC R_f 0.7 (10% MeOH/DCM). LC−MS (ESI) m/z: 287 $[M + H]^+$. ^{1}H NMR (500 MHz, $CDCl_3$) δ 7.77 (s, 1H), 7.65−7.46 (m, 2H), 7.09−6.89 (m, 2H), 5.95 (d, J = 6.2 Hz, 1H), 5.50 (br s, 2H), 3.71 (br s, 1H), 2.05 (dd, J = 12.9, 4.3 Hz, 2H), 1.78−1.83 (m, 2H), 1.67−1.71 (m, 1H), 1.47−1.33 (m, 2H), 1.22−1.30 (m, 3H). ^{13}C NMR (126 MHz, $CDCl_3$) δ 161.93, 159.61, 157.69, 135.07, 121.63, 115.39, 50.47, 32.81, 25.49, 24.88.

*4-((4-(Cyclohexylamino)pyrimidin-2-yl)amino)benzamide (**4s**).* To a mixture of **3a** (0.10 mmol, 1.0 equiv) in EtOH (0.50 mL, 0.20 M) under a nitrogen atmosphere at rt were added 4-aminobenzamide (0.20 mmol, 2.0 equiv) and a drop of 1 N HCl. The reaction mixture was heated to reflux and stirred at that temperature for 2 h; it was then cooled to rt. Purification using automated flash chromatography (MeOH/DCM) was followed by evaporation, giving **4s** as a white solid (0.011 g, 35%). TLC R_f 0.5 (10% MeOH/DCM). LC−MS (ESI) m/z: 312 $[M + H]^+$. ^{1}H NMR (400 MHz, MeOD) δ 7.87−7.79 (m, 4H), 7.75 (d, J = 6.2 Hz, 1H), 6.00 (d, J = 6.1 Hz, 1H), 3.93 (s, 1H), 2.15−2.00 (m, 2H), 1.83−1.88 (m, 2H), 1.71−1.76 (m, 1H), 1.57−1.41 (m, 2H), 1.27−1.34 (m, 3H). ^{13}C NMR (101 MHz, MeOD) δ 172.05, 163.65, 153.57, 145.07, 130.50, 129.60, 119.66, 114.60, 51.24, 33.74, 26.83, 26.36.

*N^4-Cyclohexyl-N^2-(4-(pyridin-2-yl)phenyl)pyrimidine-2,4-diamine (**4t**).* To a mixture of **3a** (0.10 mmol, 1.0 equiv) in EtOH (0.50 mL, 0.20 M) under a nitrogen atmosphere at rt were added 4-(2-pyridyl)aniline (0.11 mmol, 1.1 equiv) and a drop of 1 N HCl. The reaction mixture was heated to reflux and stirred at that temperature for 3 h; it was then cooled to rt. Purification using automated flash chromatography (EtOAc/hexanes + 2% MeOH additive) was followed by evaporation, giving **4t** as a colorless oil (0.002 g, 6%). TLC R_f 0.2 (50% EtOAc/hexanes + 1% MeOH). LC−MS (ESI) m/z: 346 $[M + H]^+$. ^{1}H NMR (500 MHz, $CDCl_3$) δ 8.59 (d, J = 4.8 Hz, 1H), 7.89 (d, J = 8.5 Hz, 2H), 7.82 (d, J = 6.2 Hz, 1H), 7.71−7.60 (m, 3H), 7.42−7.25 (m, 1H), 7.16−7.19 (m, 1H), 5.79 (d, J = 5.9 Hz, 1H), 4.76 (s, 1H), 3.84−3.45 (m, 1H), 2.07−2.10 (m, 2H), 1.67−1.79 (m, 2H), 1.66−1.56 (m, 1H), 1.45−1.30 (m, 2H), 1.21−1.29 (m, 3H). ^{13}C NMR (126 MHz, $CDCl_3$) δ 162.02, 158.96, 157.19, 149.55, 140.80, 136.63, 132.88, 127.36, 121.42, 119.86, 118.96, 118.84, 50.15, 33.06, 33.03, 25.60, 24.93.

*N^4-Cyclohexyl-N^2-(4-(pyridin-3-yl)phenyl)pyrimidine-2,4-diamine (**4u**).* To a mixture of **3a** (0.10 mmol, 1.0 equiv) in EtOH (0.50 mL, 0.20 M) under a nitrogen atmosphere at rt were added 4-pyridin-3-ylaniline (0.11 mmol, 1.1 equiv) and a drop of 1 N HCl. The reaction mixture was heated to reflux and stirred at that temperature for 4 h; it was then cooled to rt. Purification using automated flash chromatography (MeOH/DCM) was followed by evaporation, giving **4u** as a colorless oil (0.001 g, 3%). TLC R_f 0.2 (10% MeOH/DCM). LC−MS (ESI) m/z: 346 $[M + H]^+$. ^{1}H NMR (500 MHz, MeOD) δ 10.18−10.05 (m, 1H), 9.83 (d, J = 6.2 Hz, 1H), 8.96 (dd, J = 8.5, 2.1 Hz, 1H), 8.28−8.17 (m, 2H), 7.65 (d, J = 8.2 Hz, 2H), 6.88 (d, J = 8.2 Hz, 2H), 6.72 (d, J = 6.0 Hz, 1H), 4.0−4.11 (m, 1H), 2.10−2.14 (m, 2H), 1.85−1.90 (m, 2H), 1.79−1.67 (m, 1H), 1.48−1.54 (m, 2H), 1.47−1.30 (m, 3H). ^{13}C NMR (126 MHz, MeOD) δ 162.93, 155.06, 154.25, 151.12, 144.26, 141.28, 136.28, 136.14, 127.94, 127.20, 120.72, 114.97, 107.81, 49.92, 31.93, 25.34, 24.63.

*2-Chloro-N-methylpyrimidin-4-amine (**3b**).* A mixture of 2,4-dichloropyrimidine **2** (10 mmol, 1.0 equiv) and methylamine (2.0 M in MeOH) (20 mmol, 2.0 equiv) was stirred at rt in DCM (20 mL, 0.50 M) in a nitrogen atmosphere. After stirring for 16 h, the reaction mixture was concentrated to remove volatiles. Purification using automated flash chromatography (EtOAc/hexanes + 2% MeOH additive) was followed by evaporation, giving **3b** as a white solid (0.54 g, 38%). TLC R_f 0.4 (70% EtOAc/hexanes). LC−MS (ESI) m/z: 144 $[M + H]^+$. ^{1}H NMR (500 MHz, MeOD) δ 7.81 (d, J = 6.0 Hz, 1H), 6.40 (d, J = 6.4 Hz, 1H), 2.92 (s, 3H). ^{13}C NMR (126 MHz, MeOD) δ 164.37, 160.27, 153.78, 104.47, 26.17.

*N^4-Methyl-N^2-(3-(pyridin-3-yl)phenyl)pyrimidine-2,4-diamine (**5a**).* To a mixture of **3b** (0.10 mmol, 1.0 equiv) in EtOH (0.50 mL, 0.20 M) under a nitrogen atmosphere at rt were added 3-(pyridin-3-yl)aniline (0.11 mmol, 1.1 equiv) and a drop of 1 N HCl. The reaction mixture was heated to reflux and stirred at that temperature for 3 h; it was then cooled to rt. Purification using automated flash chromatography (MeOH/DCM) was followed by evaporation, giving **5a** as a colorless oil (0.006 g, 22%). TLC R_f 0.4 (10% MeOH/DCM). LC−MS (ESI) m/z: 278 $[M + H]^+$. ^{1}H NMR (500 MHz, MeOD) δ 8.89−8.77 (m, 1H), 8.52 (d, J = 5.0 Hz, 1H), 8.24−8.05 (m, 2H), 7.87−7.71 (m, 1H), 7.67 (ddd, J = 8.1, 1.8, 0.9 Hz, 1H), 7.52 (dd, J = 8.0, 4.9 Hz, 1H), 7.41 (t, J = 7.9 Hz, 1H), 7.25 (dt, J = 7.6, 1.2 Hz, 1H), 5.98 (d, J = 6.0 Hz, 1H), 2.95 (s, 3H). ^{13}C NMR (126 MHz, MeOD) δ 163.84, 159.69, 153.38, 147.22, 146.94, 141.50, 137.51, 135.13, 129.06, 124.02, 119.79, 118.90, 117.48, 97.46, 26.56.

*2-Chloro-N-ethylpyrimidin-4-amine (**3c**).* A mixture of 2,4-dichloropyrimidine **2** (10 mmol, 1.0 equiv) and ethylamine (2.0 M in THF) (20 mmol, 2.0 equiv) was stirred at rt in MeOH (10 mL, 1.0 M) in a nitrogen atmosphere. After 16 h, the reaction mixture was concentrated to remove volatiles. Purification using automated flash chromatography (EtOAc/hexanes + 2% MeOH additive) was followed by evaporation, giving **3c** as a white solid (0.43 g, 27%). TLC R_f 0.5 (70% EtOAc/hexanes). LC−MS (ESI) m/z: 160 $[M + H]^+$. ^{1}H NMR (500 MHz, $CDCl_3$) δ 8.04 (br s, 1H), 6.26 (d, J = 5.9 Hz, 1H), 5.41 (br s, 1H), 3.38 (br s, 2H), 1.28 (t, J = 7.3 Hz, 3H). ^{13}C NMR (126 MHz, $CDCl_3$) δ 163.55, 160.58, 157.26, 99.99, 36.48, 14.36.

*N^4-Ethyl-N^2-(3-(pyridin-3-yl)phenyl)pyrimidine-2,4-diamine (**5b**).* To a mixture of **3c** (0.10 mmol, 1.0 equiv) in EtOH (0.50 mL, 0.20 M) under a nitrogen atmosphere at rt were added 3-(pyridin-3-yl)aniline (0.11 mmol, 1.1 equiv) and a drop of 1 N HCl. The reaction mixture was heated to reflux and stirred at that temperature for 3 h; it was then cooled to rt. Purification using automated flash chromatography (MeOH/DCM) was followed by evaporation, giving **5b** as a colorless oil (0.003 g, 10%). TLC R_f 0.1 (5% MeOH/DCM). LC−MS (ESI) m/z: 292 $[M + H]^+$. ^{1}H NMR (500 MHz, MeOD) δ 8.84 (s, 1H), 8.55 (s, 1H), 8.18 (s, 1H), 8.13 (dt, J = 8.1, 1.8 Hz, 1H), 7.78 (s, 1H), 7.68−7.59 (m, 1H), 7.55 (dd, J = 8.0, 4.8 Hz, 1H), 7.42 (t, J = 7.9 Hz, 1H), 7.26 (dt, J = 7.7, 1.2 Hz, 1H), 5.97 (d, J = 5.4 Hz, 1H), 3.56−3.38 (m, 2H), 1.23 (t, J = 7.2 Hz, 3H). ^{13}C NMR (126 MHz, MeOD) δ 163.10, 147.24, 146.98, 141.55, 137.81, 137.57, 135.18, 129.04, 124.04, 119.79, 118.83, 117.43, 35.14, 13.49.

*2-Chloro-N-propylpyrimidin-4-amine (**3d**).* A mixture of 2,4-dichloropyrimidine **2** (10 mmol, 1.0 equiv) and cyclopentylamine (20 mmol, 2.0 equiv) was stirred at rt in DCM (20 mL, 0.50 M) in a nitrogen atmosphere. After stirring for 16 h, the reaction mixture was concentrated. Purification using automated flash chromatography (EtOAc/hexanes) was followed by evaporation, giving **3d** as a colorless oil (0.84 g, 49%). TLC R_f

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0.3 (40% EtOAc/hexanes). LC−MS (ESI) m/z: 174 $[M + H]^+$. ^{1}H NMR (500 MHz, $CDCl_3$) δ 8.04 (br s, 1H), 6.27 (d, J = 6.0 Hz, 1H), 5.54 (br s, 1H), 3.28 (br s, 2H), 1.66 (q, J = 7.3 Hz, 2H), 1.00 (t, J = 7.4 Hz, 3H). ^{13}C NMR (126 MHz, $CDCl_3$) δ 163.74, 160.55, 157.72, 99.81, 43.44, 22.35, 11.32.

*N^4-Propyl-N^2-(3-(pyridin-3-yl)phenyl)pyrimidine-2,4-diamine (**5c**).* To a mixture of **3d** (0.10 mmol, 1.0 equiv) in EtOH (0.50 mL, 0.20 M) under a nitrogen atmosphere at rt were added 3-(pyridin-3-yl)aniline (0.11 mmol, 1.1 equiv) and a drop of 1 N HCl. The reaction mixture was heated to reflux and stirred at that temperature for 3 h; it was then cooled to rt. Purification using automated flash chromatography (MeOH/DCM) was followed by evaporation, giving **5c** as a colorless oil (0.005 g, 16%). TLC R_f 0.4 (10% MeOH/DCM). LC−MS (ESI) m/z: 306 $[M + H]^+$. ^{1}H NMR (500 MHz, MeOD) δ 8.82 (s, 1H), 8.58−8.47 (m, 1H), 8.21−8.07 (m, 2H), 7.76 (s, 1H), 7.69−7.61 (m, 1H), 7.53 (dd, J = 7.9, 4.8 Hz, 1H), 7.41 (t, J = 7.9 Hz, 1H), 7.25 (dt, J = 7.7, 1.2 Hz, 1H), 5.97 (d, J = 5.8 Hz, 1H), 3.37 (s, 2H), 1.62 (q, J = 7.3 Hz, 2H), 0.92 (t, J = 7.3 Hz, 3H). ^{13}C NMR (126 MHz, MeOD) δ 163.26, 159.66, 153.40, 147.22, 147.00, 141.51, 137.76, 137.56, 135.19, 129.02, 124.02, 119.84, 118.89, 117.53, 97.60, 42.24, 22.19, 10.38.

*N-Butyl-2-chloropyrimidin-4-amine (**3e**).* To a suspension of 2,4-dichloropyrimidine **2** (10 mmol, 1.0 equiv) in DCM (20 mL, 0.50 M) was added butylamine (20 mmol, 2.0 equiv) under a nitrogen atmosphere at rt. After stirring for 16 h, the reaction mixture was concentrated. Purification using automated flash chromatography (EtOAc/hexanes) was followed by evaporation, giving **3e** as a white solid (0.96 g, 52%). TLC R_f 0.25 (30% EtOAc/hexanes). LC−MS (ESI) m/z: 188 $[M + H]^+$. ^{1}H NMR (400 MHz, $CDCl_3$) δ 8.04 (br s, 1H), 6.25 (d, J = 5.9 Hz, 1H), 5.46 (br s, 1H), 3.31 (br s, 2H), 1.71−1.51 (m, 2H), 1.51−1.33 (m, 2H), 0.97 (t, J = 7.3 Hz, 3H). ^{13}C NMR (101 MHz, $CDCl_3$) δ 163.70, 160.62, 156.38, 41.42, 31.11, 19.97, 13.71.

*N^4-Butyl-N^2-(3-(pyridin-3-yl)phenyl)pyrimidine-2,4-diamine (**5d**).* To a mixture of **3e** (0.10 mmol, 1.0 equiv) in EtOH (0.50 mL, 0.20 M) under a nitrogen atmosphere at rt were added 3-(pyridin-3-yl)aniline (0.11 mmol, 1.1 equiv) and a drop of 1 N HCl. The reaction mixture was heated to reflux and stirred at that temperature for 3 h; it was then cooled to rt. Purification using automated flash chromatography (MeOH/DCM) was followed by evaporation, giving **5d** as a colorless oil (0.006 g, 19%). TLC R_f 0.4 (10% MeOH/DCM). LC−MS (ESI) m/z: 320 $[M + H]^+$. ^{1}H NMR (400 MHz, MeOD) δ 8.70 (s, 1H), 8.48−8.33 (m, 1H), 8.07−7.95 (m, 2H), 7.63 (s, 1H), 7.53 (dd, J = 8.2, 2.1 Hz, 1H), 7.39−7.43 (m, 1H), 7.29 (t, J = 7.9 Hz, 1H), 7.13 (d, J = 7.7 Hz, 1H), 5.85 (d, J = 6.0 Hz, 1H), 3.28 (s, 2H), 1.43−1.50 (m, 2H), 1.21−1.26 (m, 2H), 0.79 (t, J = 7.4 Hz, 3H). ^{13}C NMR (101 MHz, MeOD) δ 164.65, 161.00, 148.65, 148.45, 142.88, 139.17, 136.60, 130.43, 125.42, 121.32, 120.36, 119.03, 117.61, 41.58, 32.56, 21.21, 14.19.

*2-Chloro-N-isopropylpyrimidin-4-amine (**3f**).* To a suspension of 2,4-dichloropyrimidine **2** (10 mmol, 1.0 equiv) in DCM (20 mL, 0.50 M) under a nitrogen atmosphere was added isopropylamine (20 mmol, 2.0 equiv) at rt. After stirring for 16 h, the reaction mixture was concentrated. Purification using automated flash chromatography (EtOAc/hexanes) was followed by evaporation, giving **3f** as a colorless oil (1.0 g, 60%). TLC R_f 0.10 (30% EtOAc/hexanes). LC−MS (ESI) m/z: 174 $[M + H]^+$. ^{1}H NMR (400 MHz, $CDCl_3$) δ 8.02 (s, 1H), 6.23 (d, J = 5.9 Hz, 1H), 5.10 (br s, 1H), 3.94 (br s, 1H), 1.27 (d, J = 6.5 Hz, 6H). ^{13}C NMR (126 MHz, $CDCl_3$) δ 162.81, 160.84, 157.19, 100.94, 43.25, 22.52.

*N^4-Isopropyl-N^2-(3-(pyridin-3-yl)phenyl)pyrimidine-2,4-diamine (**5e**).* To a mixture of **3f** (0.105 mmol, 1.00 equiv) in EtOH (0.500 mL, 0.210 M) under a nitrogen atmosphere at rt were added 3-(pyridin-3-yl)aniline (0.115 mmol, 1.10 equiv) and a drop of 1 N HCl. The reaction mixture was heated to reflux and stirred at that temperature for 3 h; it was then cooled to rt. Purification using automated flash chromatography (MeOH/DCM) was followed by evaporation, giving **5e** as a colorless oil (0.005 g, 16%). TLC R_f 0.5 (10% MeOH/DCM). HRMS (ESI) m/z: calcd for $[M + H]^+$ 306.1718, found 306.1720. ^{1}H NMR (500 MHz, $CDCl_3$) δ 8.87 (s, 1H), 8.59 (s, 1H), 8.05−7.82 (m, 3H), 7.55 (d, J = 5.0 Hz, 1H), 7.35−7.41 (m, 3H), 7.20 (d, J = 10.0 Hz, 1H), 5.85 (d, J = 5.7 Hz, 1H), 4.72 (s, 1H), 4.06 (s, 1H), 1.25 (d, J = 6.4 Hz, 6H). ^{13}C NMR (126 MHz, $CDCl_3$) δ 162.16, 159.77, 155.62, 148.38, 148.36, 141.01, 138.40, 136.95, 134.44, 129.41, 123.46, 120.54, 118.80, 117.78, 96.46, 42.81, 22.80.

*N-(tert-Butyl)-2-chloropyrimidin-4-amine (**3g**).* A mixture of 2,4-dichloropyrimidine **2** (10 mmol, 1.0 equiv) and *tert*-butylamine (20 mmol, 2.0 equiv) was stirred at rt in DCM (20 mL, 0.50 M) under a nitrogen atmosphere for 16 h. The reaction mixture was then concentrated to remove volatiles. Purification using automated flash chromatography (EtOAc/hexanes) was followed by evaporation, giving **3g** as a white oil (0.017 g, 1%). TLC R_f 0.3 (40% EtOAc/hexanes). LC−MS (ESI) m/z: 186 $[M + H]^+$. ^{1}H NMR (500 MHz, $CDCl_3$) δ 7.97 (d, J = 6.0 Hz, 1H), 6.25 (br s, 1H), 5.07 (br s, 1H), 1.44 (s, 9H). ^{13}C NMR (126 MHz, $CDCl_3$) δ 162.88, 160.49, 156.53, 103.33, 51.87, 28.90.

*N^4-(tert-Butyl)-N^2-(3-(pyridin-3-yl)phenyl)pyrimidine-2,4-diamine (**5f**).* To a mixture of **3g** (0.081 mmol, 1.0 equiv) in EtOH (0.50 mL, 0.16M) under a nitrogen atmosphere at rt were added 3-(pyridin-3-yl)aniline (0.089 mmol, 1.1 equiv) and a drop of 1 N HCl. The reaction mixture was heated to reflux and stirred at that temperature for 3 h; it was then cooled to rt. Purification using automated flash chromatography (MeOH/DCM) was followed by evaporation, giving **5f** as an orange oil (0.004 g, 16%). TLC R_f 0.3 (5% MeOH/DCM). LC−MS (ESI) m/z: 319 $[M + H]^+$. ^{1}H NMR (500 MHz, $CDCl_3$) δ 8.91 (br s, 1H), 8.63 (br s, 1H), 8.08−7.73 (m, 3H), 7.58 (d, J = 8.2 Hz, 1H), 7.40−7.44 (m, 3H), 7.24 (d, J = 5.0 Hz, 1H), 5.89 (d, J = 5.4 Hz, 1H), 4.85 (s, 1H), 1.46 (s, 9H). ^{13}C NMR (126 MHz, $CDCl_3$) δ 162.37, 158.85, 153.78, 148.44, 140.43, 138.58, 134.51, 129.44, 123.54, 121.10, 119.34, 118.31, 98.58, 51.59, 29.14.

*2-Chloro-N-(1-methoxypropan-2-yl)pyrimidin-4-amine (**3h**).* A mixture of 2,4-dichloropyrimidine **2** (5.0 mmol, 1.0 equiv), 1-methoxypropan-2-amine HCl (7.5 mmol, 1.5 equiv), and triethylamine (10.5 mmol, 2.1 equiv) was stirred at rt in DCM (10 mL, 0.50 M) in a nitrogen atmosphere. After 16 h, the reaction mixture was concentrated to remove volatiles. To the crude residue was added water (10 mL), and the aqueous portion was extracted into EtOAc (3 × 10 mL). The combined organics were dried over magnesium sulfate, filtered, and concentrated. Purification using automated flash chromatography (EtOAc/hexanes) was followed by evaporation, giving **3h** as a colorless oil (0.29 g, 29%). TLC R_f 0.3 (60% EtOAc/hexanes). LC−MS (ESI) m/z: 204 $[M + H]^+$. ^{1}H NMR (500 MHz, $CDCl_3$) δ 8.00 (d, J = 6.2 Hz, 1H), 6.26 (d, J = 6.0 Hz, 1H), 5.44 (br s, 1H), 4.21 (br s, 1H), 3.56−3.31 (m, 5H), 1.28 (d, J = 6.7 Hz, 3H). ^{13}C NMR (126 MHz, $CDCl_3$) δ 162.99, 160.71, 156.53, 104.44, 75.32, 59.18, 46.45, 17.43.

*N^4-(1-Methoxypropan-2-yl)-N^2-(3-(pyridin-3-yl)phenyl)-pyrimidine-2,4-diamine (**5g**).* To a mixture of **3h** (0.10 mmol, 1.0 equiv) in EtOH (0.50 mL, 0.20 M) under a nitrogen atmosphere at rt were added 3-(pyridin-3-yl)aniline (0.11 mmol,

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1.1 equiv) and a drop of 1 N HCl. The reaction mixture was heated to reflux and stirred at that temperature for 3 h; it was then cooled to rt. Purification using automated flash chromatography (MeOH/DCM) was followed by evaporation, giving **5g** as a colorless oil (0.010 g, 30%). TLC R_f 0.2 (5% MeOH/DCM). LC−MS (ESI) m/z: 336 $[M + H]^+$. ^{1}H NMR (500 MHz, MeOD) δ 8.83 (s, 1H), 8.54 (d, J = 4.9 Hz, 1H), 8.11−8.14 (m, 2H), 7.76 (d, J = 5.7 Hz, 1H), 7.63 (d, J = 5.0 Hz, 1H), 7.54 (ddd, J = 7.9, 4.9, 0.9 Hz, 1H), 7.41 (t, J = 7.9 Hz, 1H), 7.26 (d, J = 5.0 Hz, 1H), 5.99 (d, J = 6.0 Hz, 1H), 4.38 (s, 1H), 3.42 (ddd, J = 49.5, 9.4, 5.6 Hz, 2H), 3.33 (s, 3H), 1.23 (d, J = 6.7 Hz, 3H). ^{13}C NMR (126 MHz, MeOD) δ 162.75, 159.70, 153.82, 147.24, 147.00, 141.52, 137.59, 135.24, 129.04, 124.01, 119.84, 118.89, 117.51, 97.54, 75.41, 57.78, 45.46, 16.36.

*Methyl (2-chloropyrimidin-4-yl)-L-alaninate (**3i**).* A mixture of 2,4-dichloropyrimidine **2** (10 mmol, 1.0 equiv), H-Ala-OMe·HCl (10 mmol, 1.0 equiv), and triethylamine (12 mmol, 1.2 equiv) was stirred at rt in DCM (20 mL, 0.50 M) in a nitrogen atmosphere. After stirring for 16 h, the reaction mixture was concentrated to remove volatiles. To the crude residue was added water (10 mL), and the crude was extracted into EtOAc (3 × 10 mL). The combined organic phases were dried over magnesium sulfate, filtered, and concentrated. Purification using automated flash chromatography (EtOAc/hexanes) was followed by evaporation, giving **3i** as a colorless oil (0.13 g, 6%). TLC R_f 0.2 (40% EtOAc/hexanes). LC−MS (ESI) m/z: 218 $[M + H]^+$. $[\alpha]_D^{20}$ = −20 (c 0.00066, $CHCl_3$). ^{1}H NMR (500 MHz, $CDCl_3$) δ 8.15−7.97 (m, 1H), 6.33 (d, J = 5.4 Hz, 1H), 5.69 (br s, 1H), 4.73 (br s, 1H), 3.82 (s, 3H), 1.54 (d, J = 5.0 Hz, 3H). ^{13}C NMR (126 MHz, $CDCl_3$) δ 173.35, 162.39, 160.59, 160.54, 156.33, 104.28, 52.74, 49.33, 18.31.

*Methyl (2-((3-(pyridin-3-yl)phenyl)amino)pyrimidin-4-yl)-L-alaninate (**5h**).* To a mixture of **3i** (0.30 mmol, 1.0 equiv) in EtOH (1.5 mL, 0.20 M) under a nitrogen atmosphere at rt were added 3-(pyridin-3-yl)aniline (0.33 mmol, 1.1 equiv) and a drop of 1 N HCl. The reaction mixture was heated to reflux and stirred at that temperature for 3 h; it was then cooled to rt. Purification using automated flash chromatography (MeOH/DCM) was followed by evaporation, giving **5h** as an orange foam (0.024 g, 23%). TLC R_f 0.1 (5% MeOH/DCM). LC−MS (ESI) m/z: 350 $[M + H]^+$. $[\alpha]_D^{20}$ = −36 (c 0.000044, $CHCl_3$). ^{1}H NMR (500 MHz, MeOD) δ 8.85 (s, 1H), 8.54 (d, J = 4.9 Hz, 1H), 8.14 (dt, J = 8.0, 1.9 Hz, 1H), 7.97 (t, J = 2.0 Hz, 1H), 7.82 (d, J = 5.9 Hz, 1H), 7.66 (ddd, J = 8.4, 2.2, 1.0 Hz, 1H), 7.55 (dd, J = 7.9, 4.9 Hz, 1H), 7.42 (t, J = 7.9 Hz, 1H), 7.28 (ddd, J = 7.7, 1.8, 1.0 Hz, 1H), 6.09 (d, J = 5.9 Hz, 1H), 4.68 (d, J = 7.3 Hz, 1H), 3.57 (s, 3H), 1.48 (d, J = 7.3 Hz, 3H). ^{13}C NMR (126 MHz, MeOD) δ 174.42, 162.57, 159.21, 153.59, 147.25, 147.05, 141.08, 137.64, 135.25, 129.08, 124.01, 120.21, 119.23, 117.75, 97.64, 51.11, 49.18, 48.10, 16.28.

*(2-((3-(Pyridin-3-yl)phenyl)amino)pyrimidin-4-yl)-L-alanine (**5i**).* To **5h** (0.043 mmol, 1.0 equiv) in 0.72 mL (0.060 M) of THF/water (5:1) at rt under a nitrogen atmosphere was added a 1 N NaOH solution (0.13 mmol, 3.0 equiv). The reaction mixture was stirred at 80 °C for 3 h; it was then cooled to rt and concentrated to remove volatiles. The residue was diluted with water (1 mL) and then adjusted to pH 4 with 1 N HCl. The precipitated solid was filtered and washed with water, providing **5i** as a white solid (0.007 g, 49%). LC−MS (ESI) m/z: 336 $[M + H]^+$. $[\alpha]_D^{20}$ = −55 (c 0.000024, $CHCl_3$). ^{1}H NMR (500 MHz, MeOD) δ 8.81 (s, 1H), 8.54 (s, 1H), 8.16−8.06 (m, 1H), 7.72 (dd, J = 5.6, 2.8 Hz, 2H), 7.64 (d, J = 6.8 Hz, 1H), 7.54 (dd, J = 8.0, 4.9 Hz, 1H), 7.47 (t, J = 8.1 Hz, 1H), 7.37 (dt, J = 7.8, 1.4 Hz, 1H), 6.18 (d, J = 6.7 Hz, 1H), 4.51 (s, 1H), 1.49 (d, J = 7.2 Hz, 3H). ^{13}C NMR (126 MHz, MeOD) δ 176.99, 162.40, 155.33, 147.46, 146.98, 139.06, 137.96, 136.98, 135.26, 129.59, 124.10, 122.01, 120.78, 119.07, 98.50, 51.06, 16.82.

*(S)-N-(sec-Butyl)-2-chloropyrimidin-4-amine (**3j**).* To a suspension of 2,4-dichloropyrimidine **2** (2.0 mmol, 1.0 equiv) in DCM (5.0 mL, 0.40 M) under a nitrogen atmosphere was added (*S*)-(+)-2-aminobutane (4.0 mmol, 2.0 equiv) at rt. After stirring for 6 h, the reaction mixture was concentrated. Purification using automated flash chromatography (EtOAc/hexanes) was followed by evaporation, giving **3j** as a colorless oil (0.14 g, 37%). TLC R_f 0.25 (30% EtOAc/hexanes). LC−MS (ESI) m/z: 188 $[M + H]^+$. $[\alpha]_D^{20}$ = 29 (c 0.0011, $CHCl_3$). ^{1}H NMR (500 MHz, MeOD) δ 7.79 (d, J = 5.0 Hz, 1H), 6.36 (d, J = 5.0 Hz, 1H), 4.04−4.09 (m,1H), 1.64−1.49 (m, 2H), 1.19 (d, J = 6.6 Hz, 3H), 0.95 (t, J = 7.4 Hz, 3H). ^{13}C NMR (126 MHz, MeOD) δ 163.46, 160.18, 153.91, 104.38, 28.82, 18.75, 9.38.

*(S)-N^4-(sec-Butyl)-N^2-(3-(pyridin-3-yl)phenyl)pyrimidine-2,4-diamine (**5j**).* To a mixture of **3j** (0.10 mmol, 1.0 equiv) in EtOH (0.50 mL, 0.20 M) under a nitrogen atmosphere at rt were added 3-(pyridin-3-yl)aniline (0.11 mmol, 1.1 equiv) and a drop of 1 N HCl. The reaction mixture was heated to reflux and stirred at that temperature for 4 h; it was then cooled to rt. Purification using automated flash chromatography (MeOH/DCM) was followed by evaporation, giving **5j** as a yellow oil (0.007 g, 22%). TLC R_f 0.5 (10% MeOH/DCM). LC−MS (ESI) m/z: 320 $[M + H]^+$. $[\alpha]_D^{20}$ = 11 (c 0.000073, $CHCl_3$). ^{1}H NMR (500 MHz, $CDCl_3$) δ 8.86 (s, 1H), 8.58 (d, J = 4.5 Hz, 1H), 8.01−7.80 (m, 3H), 7.57 (d, J = 5.0 Hz, 1H), 7.49−7.29 (m, 3H), 7.19 (d, J = 5.8 Hz, 1H), 5.85 (d, J = 5.8 Hz, 1H), 4.77 (br s, 1H), 3.85 (br s, 1H), 1.53−1.59 (m, 2H), 1.20 (d, J = 6.4 Hz, 3H), 0.92 (t, J = 7.4 Hz, 3H). ^{13}C NMR (126 MHz, $CDCl_3$) δ 162.43, 159.48, 155.08, 148.41, 140.82, 138.47, 136.91, 134.45, 129.43, 123.46, 120.68, 118.81, 117.79, 96.43, 48.24, 29.66, 20.27, 10.36.

*(R)-N-(sec-Butyl)-2-chloropyrimidin-4-amine (**3k**).* To a suspension of 2,4-dichloropyrimidine **2** (1.0 mmol, 1.0 equiv) in DCM (3.0 mL, 0.33M) under a nitrogen atmosphere was added (*R*)-(−)-2-aminobutane (2.0 mmol, 2.0 equiv) at rt. After stirring for 6 h, the reaction mixture was concentrated. Purification using automated flash chromatography (EtOAc/hexanes) was followed by evaporation, giving **3k** as a colorless oil (0.071 g, 38%). TLC R_f 0.25 (30% EtOAc/hexanes). LC−MS (ESI) m/z: 188 $[M + H]^+$. $[\alpha]_D^{20}$ = −45 (c 0.00048, $CHCl_3$). ^{1}H NMR (500 MHz, MeOD) δ 7.79 (d, J = 6.1 Hz, 1H), 6.37 (d, J = 6.7 Hz, 1H), 4.17−3.96 (m, 1H), 1.65−1.48 (m, 2H), 1.19 (d, J = 6.6 Hz, 3H), 0.96 (t, J = 7.4 Hz, 3H). ^{13}C NMR (126 MHz, MeOD) δ 163.47, 160.19, 153.91, 104.38, 48.21, 28.81, 18.74, 9.37.

*(R)-N^4-(sec-Butyl)-N^2-(3-(pyridin-3-yl)phenyl)pyrimidine-2,4-diamine (**5k**).* To a mixture of **3k** (0.10 mmol, 1.0 equiv) in EtOH (0.50 mL, 0.20 M) under a nitrogen atmosphere at rt were added 3-(pyridin-3-yl)aniline (0.11 mmol, 1.1 equiv) and a drop of 1 N HCl. The reaction mixture was heated to reflux and stirred at that temperature for 2 h; it was then cooled to rt. Purification using automated flash chromatography (MeOH/DCM) was followed by evaporation, giving **5k** as a colorless oil (0.003 g, 9%). TLC R_f 0.3 (80% EtOAc/hexanes). LC−MS (ESI) m/z: 320 $[M + H]^+$. $[\alpha]_D^{20}$ = −10 (c 0.000033, $CHCl_3$). ^{1}H NMR (500 MHz, $CDCl_3$) δ 8.87 (s, 1H), 8.59 (s, 1H), 7.96 (s, 2H), 7.89 (dt, J = 7.8, 1.8 Hz, 1H), 7.57 (ddd, J = 8.1, 2.0, 1.0 Hz, 1H), 7.42−7.32 (m, 2H), 7.22−7.15 (m, 2H), 5.85 (d, J = 5.6 Hz, 1H), 4.64 (br s, 1H), 3.86 (br s, 1H), 1.66−1.47 (m, 2H), 1.21 (d, J = 6.5 Hz, 3H), 0.93 (t, J = 7.4 Hz, 3H). ^{13}C NMR (126 MHz,

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$CDCl_3$) δ 162.43, 159.67, 155.70, 148.44, 148.42, 140.87, 138.49, 136.94, 134.44, 129.44, 123.46, 120.63, 118.72, 117.72, 96.94, 48.20, 29.69, 20.30, 10.36.

*2-Chloro-N-cyclopropylpyrimidin-4-amine (**3l**).* A mixture of 2,4-dichloropyrimidine **2** (10 mmol, 1.0 equiv) and cyclopropylamine (10 mmol, 1.0 equiv) was stirred at rt in DCM (20 mL, 0.50 M) in a nitrogen atmosphere. After stirring for 16 h, the reaction mixture was concentrated to remove volatiles. Purification using automated flash chromatography (EtOAc/hexanes) was followed by evaporation, giving **3l** as a white solid (0.39 g, 23%). TLC R_f 0.3 (40% EtOAc/hexanes). LC−MS (ESI) m/z: 172 $[M + H]^+$. ^{1}H NMR (500 MHz, $CDCl_3$) δ 8.15 (d, J = 5.9 Hz, 1H), 6.77−6.48 (m, 1H), 5.74 (br s, 1H), 2.59 (br s, 1H), 1.00−0.83 (m, 2H), 0.72−0.54 (m, 2H). ^{13}C NMR (126 MHz, $CDCl_3$) δ 165.25, 160.30, 157.64, 101.64, 23.46, 7.71.

*N^4-Cyclopropyl-N^2-(3-(pyridin-3-yl)phenyl)pyrimidine-2,4-diamine (**5l**).* To a mixture of **3l** (0.10 mmol, 1.0 equiv) in EtOH (0.50 mL, 0.20 M) under a nitrogen atmosphere at rt were added 3-(pyridin-3-yl)aniline (0.11 mmol, 1.1 equiv) and a drop of 1 N HCl. The reaction mixture was heated to reflux and stirred at that temperature for 3 h; it was then cooled to rt. Purification using automated flash chromatography (MeOH/DCM) was followed by evaporation, giving **5l** as a colorless oil (0.004 g, 13%). TLC R_f 0.2 (5% MeOH/DCM). LC−MS (ESI) m/z: 304 $[M + H]^+$. ^{1}H NMR (500 MHz, $CDCl_3$) δ 8.87 (d, J = 2.6 Hz, 1H), 8.58 (d, J = 5.0 Hz, 1H), 8.05 (d, J = 5.0 Hz, 1H), 7.93 (s, 1H), 7.86−7.88 (m 1H), 7.56 (d, J = 8.3 Hz, 1H), 7.41−7.31 (m, 2H), 7.19 (d, J = 7.6 Hz, 1H), 7.13 (s, 1H), 6.32−6.08 (m, 1H), 5.22 (s, 1H), 2.57 (br s, 1H), 0.84−0.78 (m, 2H), 0.62−0.56 (m, 2H). ^{13}C NMR (126 MHz, $CDCl_3$) δ 164.36, 159.65, 156.84, 148.46, 148.43, 140.76, 138.48, 136.86, 134.44, 129.45, 123.49, 120.73, 118.81, 117.77, 95.85, 23.46, 7.59.

*2-Chloro-N-(1-(trifluoromethyl)cyclopropyl)pyrimidin-4-amine (**3m**).* A mixture of 2,4-dichloropyrimidine **2** (2.0 mmol, 1.0 equiv) and 1-(trifluoromethyl)cyclopropanamine (2.5 mmol, 1.25 equiv) was stirred in NMP (1.0 mL, 2.0 M) under microwave irradiation at 140 °C and 24 W for 1 h. Purification using automated flash chromatography (EtOAc/hexanes) was followed by evaporation, giving **3m** as a white solid (0.013 g, 3%). TLC R_f 0.4 (30% EtOAc/hexanes). LC−MS (ESI) m/z: 238 $[M + H]^+$. ^{1}H NMR (500 MHz, $CDCl_3$) δ 8.24 (d, J = 5.8 Hz, 1H), 6.68 (d, J = 5.8 Hz, 1H), 5.85 (br s, 1H), 1.56−1.45 (m, 2H), 1.17−1.19 (m, 2H). ^{13}C NMR (126 MHz, $CDCl_3$) δ 164.16, 160.60, 158.41, 125.15 (q, J = 277 Hz, CF_3), 119.37, 102.39, 34.46, 34.16, 33.86, 12.67, 12.66, 12.64. ^{19}F NMR (471 MHz, $CDCl_3$) δ −73.26.

*N^2-(3-(Pyridin-3-yl)phenyl)-N^4-(1-(trifluoromethyl)-cyclopropyl)pyrimidine-2,4-diamine (**5m**).* To a mixture of **3m** (0.038 mmol, 1.0 equiv) in EtOH (0.50 mL, 0.76M) under a nitrogen atmosphere at rt was added 3-(pyridin-3-yl)aniline (0.045 mmol, 1.2 equiv). The reaction mixture was heated to reflux and stirred at that temperature for 3 h; it was then cooled to rt. Purification using automated flash chromatography (MeOH/DCM) was followed by evaporation, giving **5m** as a colorless oil (0.002 g, 14%). TLC R_f 0.5 (10% MeOH/DCM). LC−MS (ESI) m/z: 372 $[M + H]^+$. ^{1}H NMR (500 MHz, $CDCl_3$) δ 8.88 (s, 1H), 8.61 (s, 1H), 8.10 (d, J = 5.8 Hz, 1H), 7.95−7.82 (m, 2H), 7.56−7.58 (m, 1H), 7.41 (t, J = 7.9 Hz, 2H), 7.25−7.18 (m, 1H), 6.99 (s, 1H), 6.21 (d, J = 5.6 Hz, 1H), 5.35 (s, 1H), 1.45−1.36 (m, 2H), 1.22−1.12 (m, 2H). ^{13}C NMR (126 MHz, $CDCl_3$) δ 163.00, 159.67, 157.42, 148.46, 140.39, 138.63, 134.45, 129.50, 125.46 (q, J = 275.94, CF_3), 122.16, 121.12, 118.96, 117.91, 96.47, 34.56, 34.25, 33.95, 33.65, 12.74. ^{19}F NMR (471 MHz, $CDCl_3$) δ −73.18.

*2-Chloro-N-cyclobutylpyrimidin-4-amine (**3n**).* A mixture of 2,4-dichloropyrimidine **2** (10 mmol, 1.0 equiv) and cyclobutylamine (10 mmol, 1.0 equiv) was stirred at rt in DCM (20 mL, 0.50 M) in a nitrogen atmosphere. After stirring for 16 h, the reaction mixture was concentrated to remove volatiles. Purification using automated flash chromatography (EtOAc/hexanes) was followed by evaporation, giving **3n** as a white solid (0.059 g, 3%). TLC R_f 0.3 (40% EtOAc/hexanes). LC−MS (ESI) m/z: 184 $[M + H]^+$. ^{1}H NMR (500 MHz, $CDCl_3$) δ 8.04 (d, J = 10.9 Hz, 1H), 6.21 (d, J = 10.0 Hz, 1H), 5.48 (br s, 1H), 4.01 (br s, 1H), 2.57−2.40 (m, 2H), 1.92−1.97 (m, 2H), 1.90−1.74 (m, 2H). ^{13}C NMR (126 MHz, $CDCl_3$) δ 162.57, 160.51, 157.78, 100.35, 46.73, 31.27, 30.75, 15.16.

*N^4-Cyclobutyl-N^2-(3-(pyridin-3-yl)phenyl)pyrimidine-2,4-diamine (**5n**).* To a mixture of **3n** (0.10 mmol, 1.0 equiv) in EtOH (0.50 mL, 0.20 M) under a nitrogen atmosphere at rt were added 3-(pyridin-3-yl)aniline (0.11 mmol, 1.1 equiv) and a drop of 1 N HCl. The reaction mixture was heated to reflux and stirred at that temperature for 3 h; it was then cooled to rt. Purification using automated flash chromatography (MeOH/DCM) was followed by evaporation, giving **5n** as a yellow oil (0.002 g, 6%). TLC R_f 0.1 (5% MeOH/DCM). LC−MS (ESI) m/z: 318 $[M + H]^+$. ^{1}H NMR (500 MHz, MeOD) δ 8.85 (s, 1H), 8.62−8.49 (m, 1H), 8.35−8.14 (m, 1H), 8.13 (dt, J = 8.0, 1.9 Hz, 1H), 7.77 (s, 1H), 7.54−7.60 (m, 2H), 7.42 (t, J = 7.9 Hz, 1H), 7.26 (d, J = 7.6, 1H), 5.94 (d, J = 5.9 Hz, 1H), 4.40−4.56 (m, 1H), 2.42−2.25 (m, 2H), 1.92−2.01 (m, 2H), 1.79−1.54 (m, 2H). ^{13}C NMR (126 MHz, MeOD) δ 162.15, 159.43, 153.31, 147.28, 147.04, 141.37, 137.82, 137.63, 135.20, 129.03, 124.01, 120.00, 118.93, 117.64, 97.37, 45.88, 30.32, 14.57.

*2-Chloro-N-cyclopentylpyrimidin-4-amine (**3o**).* A mixture of 2,4-dichloropyrimidine **2** (10 mmol, 1.0 equiv) and cyclopentylamine (20 mmol, 2.0 equiv) was stirred at rt in DCM (20 mL, 0.50 M) in a nitrogen atmosphere. After 16 h, the reaction mixture was concentrated. Purification using automated flash chromatography (EtOAc/hexanes) followed by evaporation gave **3o** as a yellow oil (1.12 g, 57%). TLC R_f 0.25 (40% EtOAc/hexanes). LC−MS (ESI) m/z: 200 $[M + H]^+$. ^{1}H NMR (500 MHz, $CDCl_3$) δ 8.03 (d, J = 11.7 Hz, 1H), 6.32 (d, J = 6.3 Hz, 1H), 5.46 (br s, 1H), 3.93 (br s, 1H), 2.19−1.94 (m, 2H), 1.87−1.63 (m, 4H), 1.51−1.56 (m, 2H). ^{13}C NMR (126 MHz, $CDCl_3$) δ 163.22, 160.56, 157.25, 100.46, 53.12, 33.08, 23.66.

*N^4-Cyclopentyl-N^2-(3-(pyridin-3-yl)phenyl)pyrimidine-2,4-diamine (**5o**).* To a mixture of **3o** (0.10 mmol, 1.0 equiv) in EtOH (0.50 mL, 0.20 M) under a nitrogen atmosphere at rt were added 3-(pyridin-3-yl)aniline (0.11 mmol, 1.1 equiv) and a drop of 1 N HCl. The reaction mixture was heated to reflux and stirred at that temperature for 3 h; it was then cooled to rt. Purification using automated flash chromatography (MeOH/DCM) was followed by evaporation, giving **5o** as a white oil (0.005 g, 15%). TLC R_f 0.5 (10% MeOH/DCM). LC−MS (ESI) m/z: 332 $[M + H]^+$. ^{1}H NMR (400 MHz, MeOD) δ 8.81 (s, 1H), 8.53 (d, J = 5.1 Hz, 1H), 8.21 (br s, 1H), 8.09 (d, J = 7.5 Hz, 1H), 7.74 (s, 1H), 7.67−7.56 (m, 1H), 7.51−7.54 (m, 1H), 7.40 (t, J = 7.9 Hz, 1H), 7.23 (dd, J = 7.6, 1.9 Hz, 1H), 5.96 (d, J = 5.8 Hz, 1H), 4.30 (s, 1H), 1.96−2.01 (m, 2H), 1.68−1.75 (m, 2H), 1.45−1.53 (m, 4H). ^{13}C NMR (101 MHz, MeOD) δ 164.22, 161.02, 148.64, 148.46, 142.94, 139.27, 139.02, 136.62, 130.40, 125.39, 121.27, 120.25, 118.99, 53.54, 33.77, 24.70.

*N-(2-Chloropyrimidin-4-yl)-O-methylhydroxylamine (**3p**).* A mixture of 2,4-dichloropyrimidine **2** (10 mmol, 1.0 equiv),

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O-methylhydroxylamine HCl (15 mmol, 1.5 equiv), and triethylamine (20 mmol, 2.0 equiv) was stirred at rt in a 0.50 M mixture of DCM (10 mL) and MeOH (10 mL) under a nitrogen atmosphere for 16 h. The reaction mixture was then concentrated to remove volatiles. To the crude residue was added water (10 mL), and then, the crude was extracted into EtOAc (3 × 10 mL). The organic phase was dried over magnesium sulfate, filtered, and concentrated. Purification using automated flash chromatography (EtOAc/hexanes) was followed by evaporation, giving **3p** as a white solid (0.19 g, 12%). TLC R_f 0.5 (50% EtOAc/hexanes). LC−MS (ESI) *m*/*z*: 162 $[M + H]^+$. ^{1}H NMR (500 MHz, $CDCl_3$) δ 8.47 (br s, 1H), 8.31 (d, *J* = 5.7 Hz, 1H), 6.78 (d, *J* = 5.8 Hz, 1H), 3.84 (s, 3H). ^{13}C NMR (126 MHz, $CDCl_3$) δ 166.53, 159.99, 158.94, 101.72, 64.25.

*4-(Methoxyamino)-N-(3-(pyridin-3-yl)phenyl)pyrimidin-2-amine (**5p**).* To a mixture of **3p** (0.10 mmol, 1.0 equiv) in EtOH (0.50 mL, 0.20 M) under a nitrogen atmosphere at rt were added 3-(pyridin-3-yl)aniline (0.11 mmol, 1.1 equiv) and a drop of 1 N HCl. The reaction mixture was heated to reflux and stirred at that temperature for 3 h; it was then cooled to rt. Purification using automated flash chromatography (MeOH/DCM) was followed by evaporation, giving **5p** as a colorless oil (0.006 g, 20%). TLC R_f 0.4 (5% MeOH/DCM). LC−MS (ESI) *m*/*z*: 294 $[M + H]^+$. ^{1}H NMR (500 MHz, $CDCl_3$) δ 8.90 (s, 1H), 8.63 (d, *J* = 5.0 Hz, 1H), 8.24 (d, *J* = 5.6 Hz, 1H), 7.98−7.86 (m, 2H), 7.61 (s, 1H), 7.56 (d, *J* = 8.2 Hz, 1H), 7.46−7.34 (m, 2H), 7.27−7.21 (m, 1H), 7.18−7.09 (m, 1H), 6.40 (d, *J* = 5.6 Hz, 1H), 3.83 (s, 3H). ^{13}C NMR (126 MHz, $CDCl_3$) δ 166.38, 159.30, 158.40, 148.49, 148.41, 140.30, 138.54, 136.70, 134.44, 129.54, 123.53, 121.14, 118.95, 117.94, 95.89, 64.15.

*tert-Butyl 4-((2-chloropyrimidin-4-yl)amino)piperidine-1-carboxylate (**3q**).* To a suspension of 2,4-dichloropyrimidine **2** (5.0 mmol, 1.0 equiv) in DCM (10 mL, 0.50 M) under a nitrogen atmosphere was added 1-Boc-4-aminopiperidine (7.5 mmol, 1.5 equiv) at rt. After stirring for 5 h the reaction mixture was concentrated. Purification using automated flash chromatography (EtOAc/hexanes) was followed by evaporation, giving **3q** as a white foam (0.41 g, 26%). TLC R_f 0.10 (30% EtOAc/hexanes). LC−MS (ESI) *m*/*z*: 313 $[M + H]^+$. ^{1}H NMR (400 MHz, MeOD) δ 7.85 (br s, 1H), 6.40 (br s, 1H), 4.03−4.13 (m, 3H), 2.99 (s, 2H), 2.06−1.89 (m, 2H), 1.48 (s, 9H), 1.45−1.35 (m, 2H). ^{13}C NMR (101 MHz, MeOD) δ 164.55, 161.59, 156.45, 155.77, 105.96, 101.41, 81.17, 43.88, 32.45, 28.68.

*tert-Butyl 4-((2-((3-(pyridin-3-yl)phenyl)amino)pyrimidin-4-yl)amino)piperidine-1-carboxylate (**3r**).* To a mixture of **3q** (0.10 mmol, 1.0 equiv) in EtOH (0.50 mL, 0.20 M) under a nitrogen atmosphere at rt were added 3-(pyridin-3-yl)aniline (0.11 mmol, 1.1 equiv) and a drop of 1 N HCl. The reaction mixture was heated to reflux and stirred at that temperature for 3 h; it was then cooled to rt. Purification using automated flash chromatography (MeOH/DCM) was followed by evaporation, giving **3r** as a colorless oil (0.007 g, 16%). TLC R_f 0.5 (10% MeOH/DCM). LC−MS (ESI) *m*/*z*: 447 $[M + H]^+$. ^{1}H NMR (500 MHz, MeOD) δ 8.82 (s, 1H), 8.53 (d, *J* = 5.0 Hz, 1H), 8.19−8.05 (m, 2H), 7.77 (s, 1H), 7.59 (d, *J* = 7.4 Hz, 1H), 7.54 (dd, *J* = 7.9, 4.9 Hz, 1H), 7.42 (t, *J* = 7.9 Hz, 1H), 7.31−7.16 (m, 1H), 5.97 (d, *J* = 5.9 Hz, 1H), 4.02 (s, 1H), 3.98−3.84 (m, 2H), 2.60−2.69 (m, 2H), 1.99−1.90 (m, 2H), 1.47 (s, 9H), 1.40−1.28 (m, 2H). ^{13}C NMR (126 MHz, MeOD) δ 162.35, 159.45, 154.98, 153.44, 147.35, 147.11, 141.31, 137.87, 137.60, 135.29, 129.05, 124.10, 120.17, 119.14, 117.92, 97.70, 79.69, 53.40, 42.05, 31.26, 27.27.

*N^4-(Piperidin-4-yl)-N^2-(3-(pyridin-3-yl)phenyl)pyrimidine-2,4-diamine (**5q**).* To **3r** (0.020 mmol, 1.0 equiv) in DCM (1.0 mL, 0.20 M) under a nitrogen atmosphere at rt was added trifluoroacetic acid (0.20 mmol, 10 equiv). The reaction mixture was stirred at rt for 3 h and then concentrated. Purification using automated reversed-phase flash chromatography (water/MeOH) was followed by evaporation, giving **5q** as a colorless oil (0.007 g, 100%). LC−MS (ESI) *m*/*z*: 347 $[M + H]^+$. ^{1}H NMR (500 MHz, MeOD) δ 8.91 (s, 1H), 8.63 (s, 1H), 8.16 (d, *J* = 7.9 Hz, 1H), 8.02 (s, 1H), 7.80 (s, 1H), 7.61 (d, *J* = 8.3 Hz, 2H), 7.52 (t, *J* = 7.8 Hz, 1H), 7.43 (d, *J* = 7.7 Hz, 1H), 6.15 (d, *J* = 6.2 Hz, 1H), 4.16−4.20 (m, 1H), 3.35−3.39 (m, 2H), 3.00−2.81 (m, 2H), 2.22−2.26 (m, 2H), 1.84−1.70 (m, 2H). ^{13}C NMR (126 MHz, MeOD) δ 165.13, 162.56, 161.59, 156.29, 147.68, 147.00, 139.44, 137.92, 135.19, 129.43, 124.45, 122.05, 120.74, 119.38, 98.25, 45.38, 42.46, 27.85.

*2-Chloro-N-(tetrahydro-2H-pyran-4-yl)pyrimidin-4-amine (**3s**).* To a suspension of 2,4-dichloropyrimidine **2** (2.5 mmol, 1.0 equiv) in DCM (5.0 mL, 0.50 M) under a nitrogen atmosphere was added 4-aminotetrohydropyran (5.0 mmol, 2.0 equiv) at rt. After stirring for 16 h, the reaction mixture was concentrated. Purification using automated flash chromatography (EtOAc/hexanes) was followed by evaporation, giving **3s** as a white solid (0.19 g, 35%). TLC R_f 0.30 (70% EtOAc/hexanes). LC−MS (ESI) *m*/*z*: 216 $[M + H]^+$. ^{1}H NMR (500 MHz, $CDCl_3$) δ 8.12−7.94 (m, 1H), 6.25 (d, *J* = 5.9 Hz, 1H), 5.01 (br s, 1H), 4.00−4.04 (m, 2H), 3.53−3.59 (m, 2H), 2.11−1.96 (m, 2H), 1.63−1.49 (m, 2H). ^{13}C NMR (126 MHz, $CDCl_3$) δ 162.63, 160.99, 157.11, 66.47, 47.24, 32.83.

*N^2-(3-(Pyridin-3-yl)phenyl)-N^4-(tetrahydro-2H-pyran-4-yl)-pyrimidine-2,4-diamine (**5r**).* To a mixture of **3s** (0.094 mmol, 1.0 equiv) in EtOH (0.50 mL, 0.19 M) under a nitrogen atmosphere at rt were added 3-(pyridin-3-yl)aniline (0.103 mmol, 1.1 equiv) and a drop of 1 N HCl. The reaction mixture was heated to reflux and stirred at that temperature for 4 h; it was then cooled to rt. Purification using automated flash chromatography (MeOH/DCM) was followed by evaporation, giving **5r** as a colorless oil (0.005 g, 15%). TLC R_f 0.6 (10% MeOH/DCM). LC−MS (ESI) *m*/*z*: 348 $[M + H]^+$. ^{1}H NMR (500 MHz, $CDCl_3$) δ 8.80 (s, 1H), 8.53 (d, *J* = 5.1 Hz, 1H), 7.94−7.84 (m, 2H), 7.82 (d, *J* = 8.0 Hz, 1H), 7.53−7.43 (m, 1H), 7.35−7.27 (m, 2H), 7.23 (s, 1H), 7.13 (dd, *J* = 7.7, 1.6 Hz, 1H), 5.80 (d, *J* = 5.8 Hz, 1H), 4.66 (s, 1H), 3.91 (br s, 1H), 3.86 (d, *J* = 11.5 Hz, 2H), 3.28 (t, *J* = 11.7 Hz, 2H), 1.93 (d, *J* = 10.0 Hz, 2H), 1.54−1.35 (m, 2H). ^{13}C NMR (126 MHz, $CDCl_3$) δ 161.92, 159.68, 155.79, 148.51, 148.43, 140.74, 138.52, 136.95, 134.46, 129.43, 123.53, 120.84, 118.81, 117.87, 97.05, 66.64, 47.19, 33.17.

*N^2-([1,1′-Biphenyl]-2-yl)-N^4-isopropylpyrimidine-2,4-diamine (**6a**).* To a mixture of **3f** (0.10 mmol, 1.0 equiv) in EtOH (0.50 mL, 0.20 M) under a nitrogen atmosphere at rt were added 2-aminobiphenyl (0.15 mmol, 1.5 equiv) and a drop of 1 N HCl. The reaction mixture was heated to reflux and stirred at that temperature for 2 h; it was then cooled to rt. Purification using automated flash chromatography (MeOH/DCM) was followed by evaporation, giving **6a** as a white solid (0.021 g, 69%). TLC R_f 0.7 (10% MeOH/DCM). LC−MS (ESI) *m*/*z*: 305 $[M + H]^+$. ^{1}H NMR (400 MHz, $CDCl_3$) δ 8.22 (d, *J* = 8.2 Hz, 1H), 7.71 (d, *J* = 5.9 Hz, 1H), 7.40−7.33 (m, 4H), 7.31−7.24 (m, 2H), 7.17 (d, *J* = 1.7 Hz, 1H), 7.03 (td, *J* = 7.4, 1.2 Hz, 1H), 5.73 (d, *J* = 6.0 Hz, 1H), 4.84 (br s, 1H), 3.86 (br s, 1H), 1.15 (d, *J* = 6.4 Hz, 6H). ^{13}C NMR (101 MHz, $CDCl_3$) δ 162.03, 158.76, 138.82, 136.31, 132.73, 130.26, 129.40, 128.90, 127.89, 127.58, 122.79, 121.39, 42.97, 22.72.

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*N^2-(3-(tert-Butyl)phenyl)-N^4-isopropylpyrimidine-2,4-diamine (**6b**).* To a mixture of **3f** (0.15 mmol, 1.0 equiv) in EtOH (0.50 mL, 0.30 M) under a nitrogen atmosphere at rt were added 3-(*tert*-butyl)aniline (0.22 mmol, 1.5 equiv) and a drop of 1 N HCl. The reaction mixture was heated to reflux and stirred at that temperature for 1 h; it was then cooled to rt. Purification using automated flash chromatography (MeOH/DCM) was followed by evaporation, giving **6b** as a colorless oil (0.027 g, 65%). TLC R_f 0.6 (10% MeOH/DCM). LC−MS (ESI) m/z: 285 $[M + H]^+$. ^{1}H NMR (500 MHz, $CDCl_3$) δ 8.74 (br s, 1H), 7.63 (d, J = 31.1 Hz, 2H), 7.47 (d, J = 8.0 Hz, 1H), 7.35−7.22 (m, 2H), 7.14 (d, J = 7.9 Hz, 1H), 6.05 (d, J = 6.5 Hz, 1H), 4.23 (br s, 1H), 1.35 (d, J = 2.0 Hz, 9H), 1.30 (d, J = 8.0 Hz, 6H). ^{13}C NMR (126 MHz, $CDCl_3$) δ 161.95, 152.02, 137.97, 128.41, 120.65, 117.70, 117.59, 43.36, 34.77, 31.32, 22.53.

*N^2-([1,1′-Biphenyl]-3-yl)-N^4-isopropylpyrimidine-2,4-diamine (**6c**).* To a mixture of **3f** (0.10 mmol, 1.0 equiv) in EtOH (0.50 mL, 0.20 M) under a nitrogen atmosphere at rt were added 3-aminobiphenyl (0.15 mmol, 1.5 equiv) and a drop of 1 N HCl. The reaction mixture was heated to reflux and stirred at that temperature for 2 h; it was then cooled to rt. Purification using automated flash chromatography (MeOH/DCM) was followed by evaporation, giving **6c** as a gray solid (0.020 g, 66%). TLC R_f 0.7 (10% MeOH/DCM). LC−MS (ESI) m/z: 305 $[M + H]^+$. ^{1}H NMR (400 MHz, chloroform-*d*) δ 7.81 (d, J = 8.3 Hz, 2H), 7.56−7.47 (m, 3H), 7.43 (d, J = 11.1 Hz, 1H), 7.35 (td, J = 8.2, 7.7, 1.7 Hz, 2H), 7.31−7.23 (m, 2H), 7.15 (dt, J = 7.7, 1.5 Hz, 1H), 5.76 (dd, J = 6.0, 1.3 Hz, 1H), 4.73 (br s, 1H), 4.01 (br s, 1H), 1.17 (d, J = 8.0 Hz, 6H). ^{13}C NMR (101 MHz, $CDCl_3$) δ 162.12, 159.25, 141.92, 141.38, 140.27, 129.09, 128.62, 127.23, 121.03, 118.25, 118.12, 42.89, 22.80.

*N^4-Isopropyl-N^2-(3-(pyridin-2-yl)phenyl)pyrimidine-2,4-diamine (**6d**).* To a mixture of **3f** (0.14 mmol, 1.0 equiv) in EtOH (0.50 mL, 0.28 M) under a nitrogen atmosphere at rt were added 3-(2-pyridyl)aniline (0.15 mmol, 1.1 equiv) and a drop of 1 N HCl. The reaction mixture was heated to reflux and stirred at that temperature for 3 h; it was then cooled to rt. Purification using automated flash chromatography (MeOH/DCM) was followed by evaporation, giving **6d** as a colorless oil (0.014 g, 33%). TLC R_f 0.5 (10% MeOH/DCM). LC−MS (ESI) m/z: 306 $[M + H]^+$. ^{1}H NMR (400 MHz, MeOD) δ 8.64 (d, J = 4.9 Hz, 1H), 8.36 (s, 1H), 7.99−7.83 (m, 2H), 7.67−7.70 (m, 2H), 7.64−7.54 (m, 1H), 7.49 (t, J = 7.9 Hz, 1H), 7.45−7.31 (m, 1H), 6.09 (d, J = 6.7 Hz, 1H), 4.33 (br s, 1H), 1.24 (d, J = 6.5 Hz, 6H). ^{13}C NMR (101 MHz, MeOD) δ 163.57, 158.73, 150.33, 141.35, 140.40, 138.91, 130.44, 123.95, 123.43, 122.73, 122.69, 120.88, 101.41, 44.04, 22.46.

*N^4-Isopropyl-N^2-(3′-methoxy-[1,1′-biphenyl]-3-yl)-pyrimidine-2,4-diamine (**6e**).* To a mixture of **3f** (0.070 mmol, 1.0 equiv) in EtOH (0.50 mL, 0.14 M) under a nitrogen atmosphere at rt were added (3′-methoxybiphenyl-3-yl)amine HCl salt (0.084 mmol, 1.2 equiv) and a drop of 1 N HCl. The reaction mixture was heated to reflux and stirred at that temperature for 16 h; it was then cooled to rt. Purification using automated flash chromatography (MeOH/DCM) was followed by evaporation, giving **6e** as a colorless oil (0.013 g, 56%). TLC R_f 0.2 (50% EtOAc/hexanes + 1% MeOH). LC−MS (ESI) m/z: 335 $[M + H]^+$. ^{1}H NMR (400 MHz, $CDCl_3$) δ 8.09−7.88 (m, 2H), 7.74−7.63 (m, 2H), 7.53−7.40 (m, 2H), 7.40−7.27 (m, 3H), 7.07−6.92 (m, 1H), 5.96 (d, J = 8.0 Hz, 1H), 4.95 (d, J = 7.8 Hz, 1H), 4.21 (br s, 1H), 3.89 (s, 3H), 1.36 (d, J= 8.0 Hz, 6H). ^{13}C NMR (126 MHz, $CDCl_3$) δ 162.10, 159.86, 159.03, 146.68, 142.90, 141.79, 140.14, 129.60, 129.08, 121.13, 119.76, 118.45, 118.20, 113.08, 112.58, 96.25, 55.31, 42.97, 22.76.

*Methyl 3′-((4-(isopropylamino)pyrimidin-2-yl)amino)-[1,1′-biphenyl]-3-carboxylate (**6f**).* To a mixture of **3f** (0.12 mmol, 1.0 equiv) in MeOH (0.50 mL, 0.23 M) under a nitrogen atmosphere at rt were added 3′-amino-biphenyl-3-carboxylic acid methyl ester HCl (0.13 mmol, 1.1 equiv) and a drop of 1 N HCl. The reaction mixture was heated to reflux and stirred at that temperature for 3 h; it was then cooled to rt. The reaction mixture was diluted with EtOAc (3 mL) and then washed sequentially with 1 N HCl (3 mL) and brine (3 mL). The organic phase was dried over magnesium sulfate and concentrated. Purification using automated flash chromatography (MeOH/DCM) was followed by evaporation, giving **6f** as a white solid (0.015 g, 36%). TLC R_f 0.6 (10% MeOH/DCM). LC−MS (ESI) m/z: 363 $[M + H]^+$. ^{1}H NMR (400 MHz, $CDCl_3$) δ 8.30 (d, J = 1.9 Hz, 1H), 8.05−7.87 (m, 3H), 7.80 (dt, J = 8.0, 1.4 Hz, 1H), 7.62−7.52 (m, 1H), 7.50 (t, J = 7.7 Hz, 1H), 7.38 (t, J = 7.9 Hz, 1H), 7.26−7.19 (m, 1H), 7.09 (s, 1H), 5.83 (d, J = 5.8 Hz, 1H), 4.63 (br s, 1H), 4.05 (br s, 1H), 3.95 (s, 3H), 1.24 (d, J = 6.4 Hz, 6H). ^{13}C NMR (101 MHz, $CDCl_3$) δ 167.08, 162.14, 159.85, 155.96, 141.74, 140.80, 140.73, 131.64, 130.58, 129.24, 128.70, 128.38, 128.31, 120.68, 118.40, 117.81, 52.17, 42.81, 22.82.

*N^4-Isopropyl-N^2-(3-(oxazol-2-yl)phenyl)pyrimidine-2,4-diamine (**6g**).* To a mixture of **3f** (0.10 mmol, 1.0 equiv) in EtOH (0.50 mL, 0.20 M) under a nitrogen atmosphere at rt were added 3-(1,3-oxaxol-2-yl)aniline (0.11 mmol, 1.1 equiv) and a drop of 1 N HCl. The reaction mixture was heated to reflux and stirred at that temperature for 3 h; it was then cooled to rt. Purification using automated flash chromatography (MeOH/DCM) was followed by evaporation, giving **6g** as a colorless oil (0.005 g, 17%). TLC R_f 0.5 (10% MeOH/DCM). LC−MS (ESI) m/z: 296 $[M + H]^+$. ^{1}H NMR (500 MHz, MeOD) δ 8.66 (t, J = 2.0 Hz, 1H), 7.99 (d, J = 0.9 Hz, 1H), 7.88−7.69 (m, 1H), 7.69−7.57 (m, 2H), 7.40 (t, J = 7.9 Hz, 1H), 7.31 (d, J = 0.9 Hz, 1H), 5.96 (d, J = 6.0 Hz, 1H), 4.33 (br s, 1H), 1.27 (d, J = 6.5 Hz, 6H). ^{13}C NMR (126 MHz, MeOD) δ 162.52, 162.41, 159.55, 153.55, 141.51, 139.19, 128.77, 127.53, 127.10, 120.97, 118.86, 116.51, 97.57, 41.90, 21.44.

*N^4-Isopropyl-N^2-(3-(thiazol-2-yl)phenyl)pyrimidine-2,4-diamine (**6h**).* To a mixture of **3f** (0.10 mmol, 1.0 equiv) in EtOH (0.50 mL, 0.20 M) under a nitrogen atmosphere at rt were added 3-(1,3-thiazol-2-yl)aniline (0.11 mmol, 1.1 equiv) and a drop of 1 N HCl. The reaction mixture was heated to reflux and stirred at that temperature for 3 h; it was then cooled to rt. Purification using automated flash chromatography (MeOH/DCM) was followed by evaporation, giving **6h** as a colorless oil (0.004 g, 13%). TLC R_f 0.5 (10% MeOH/DCM). LC−MS (ESI) m/z: 312 $[M + H]^+$. ^{1}H NMR (500 MHz, MeOD) δ 8.53 (t, J = 2.1 Hz, 1H), 7.87 (d, J = 3.3 Hz, 1H), 7.83−7.68 (m, 1H), 7.68−7.58 (m, 2H), 7.54 (d, J = 10.0 Hz, 1H), 7.38 (t, J = 7.9 Hz, 1H), 5.96 (d, J = 6.0 Hz, 1H), 4.35 (br s, 1H), 1.25 (d, J = 6.5 Hz, 6H). ^{13}C NMR (126 MHz, MeOD) δ 169.29, 162.43, 159.55, 153.50, 142.90, 141.60, 133.43, 128.90, 120.62, 119.18, 116.81, 97.61, 41.85, 21.44.

*N^2-(3-(1H-Pyrazol-3-yl)phenyl)-N^4-isopropylpyrimidine-2,4-diamine (**6i**).* To a mixture of **3f** (0.10 mmol, 1.0 equiv) in EtOH (0.50 mL, 0.20 M) under a nitrogen atmosphere at rt were added 3-(1*H*-pyrazol-3-yl)aniline (0.11 mmol, 1.1 equiv) and a drop of 1 N HCl. The reaction mixture was heated to reflux and stirred at that temperature for 3 h; it was then cooled to rt. Purification using automated flash chromatography (MeOH/DCM) was followed by evaporation, giving **6i** as a white oil

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(0.020 g, 68%). TLC R_f 0.3 (10% MeOH/DCM). LC−MS (ESI) m/z: 295 $[M + H]^+$. ^1H NMR (500 MHz, MeOD) δ 8.12 (br s, 1H), 7.81−7.71 (m, 1H), 7.67 (br s, 1H), 7.61−7.54 (m, 1H), 7.31−7.38 (m, 2H), 6.65 (br s, 1H), 5.94 (d, J = 6.1 Hz, 1H), 4.26 (br s, 1H), 1.24 (d, J = 6.5 Hz, 6H). ^{13}C NMR (126 MHz, MeOD) δ 162.44, 159.53, 153.19, 151.87, 140.94, 133.84, 129.47, 128.57, 118.84, 116.64, 101.95, 97.35.

*N^2-(3-(1H-Imidazol-1-yl)phenyl)-N^4-isopropylpyrimidine-2,4-diamine (**6j**).* To a mixture of **3f** (0.10 mmol, 1.0 equiv) in EtOH (0.50 mL, 0.20 M) under a nitrogen atmosphere at rt were added 3-(1*H*-imidazol-1-yl)aniline (0.11 mmol, 1.1 equiv) and a drop of 1 N HCl. The reaction mixture was heated to reflux and stirred at that temperature for 6 h; it was then cooled to rt. Purification using automated flash chromatography (MeOH/DCM) and then reversed-phase chromatography (MeCN/water) was followed by evaporation, giving **6j** as a colorless oil (0.004 g, 14%). LC−MS (ESI) m/z: 295 $[M + H]^+$. ^1H NMR (500 MHz, MeOD) δ 8.28 (br s, 1H), 8.15 (br s, 1H), 7.75 (br s, 2H), 7.54−7.56 (m, 1H), 7.48 (t, J = 8.0 Hz, 1H), 7.45−7.09 (m, 2H), 6.07 (d, J = 5.8 Hz, 1H), 4.24 (br s, 1H), 1.25 (d, J = 6.5 Hz, 6H). ^{13}C NMR (126 MHz, MeOD) δ 162.22, 156.66, 147.86, 140.93, 137.76, 129.90, 118.82, 115.17, 112.80, 98.35, 42.50, 21.05.

*N^4-Isopropyl-N^2-(3-(pyrazin-2-yl)phenyl)pyrimidine-2,4-diamine (**6k**).* To a mixture of **3f** (0.10 mmol, 1.0 equiv) in EtOH (0.50 mL, 0.20 M) under a nitrogen atmosphere at rt were added 3-(pyrazin-2-yl)aniline (0.11 mmol, 1.1 equiv) and a drop of 1 N HCl. The reaction mixture was heated to reflux and stirred at that temperature for 3 h; it was then cooled to rt. Purification using automated flash chromatography (MeOH/DCM) was followed by evaporation, giving **6k** as a colorless foam (0.011 g, 36%). TLC R_f 0.5 (10% MeOH/DCM). LC−MS (ESI) m/z: 307 $[M + H]^+$. ^1H NMR (500 MHz, MeOD) δ 9.09 (d, J = 1.5 Hz, 1H), 8.68 (dd, J = 2.6, 1.6 Hz, 1H), 8.63 (s, 1H), 7.74 (br s, 1H), 7.70−7.61 (m, 2H), 7.43 (t, J = 7.9 Hz, 1H), 5.95 (d, J = 6.0 Hz, 1H), 4.45−4.17 (m, 1H), 1.25 (d, J = 6.5 Hz, 6H). ^{13}C NMR (126 MHz, MeOD) δ 162.41, 159.46, 153.34, 144.14, 142.44, 141.78, 141.45, 136.42, 128.86, 120.48, 119.72, 117.46, 97.33, 41.84, 21.43.

*N^4-isopropyl-N^2-(3-(pyridin-4-yl)phenyl)pyrimidine-2,4-diamine (**6l**).* To a mixture of **3f** (0.10 mmol, 1.0 equiv) in EtOH (0.50 mL, 0.20 M) under a nitrogen atmosphere at rt were added 3-(4-pyridyl)aniline (0.11 mmol, 1.1 equiv) and a drop of 1 N HCl. The reaction mixture was heated to reflux and stirred at that temperature for 3 h; it was then cooled to rt. Purification using automated flash chromatography (MeOH/DCM) was followed by evaporation, giving **6l** as an orange solid (0.002 g, 7%). TLC R_f 0.5 (10% MeOH/DCM). LC−MS (ESI) m/z: 306 $[M + H]^+$. ^1H NMR (500 MHz, $CDCl_3$) δ 8.74−8.59 (m, 2H), 8.07 (br s, 1H), 7.95 (d, J = 5.7 Hz, 1H), 7.64−7.58 (m, 1H), 7.58−7.51 (m, 2H), 7.43 (t, J = 7.9 Hz, 1H), 7.27 (s, 1H), 7.14 (br s, 1H), 5.88 (d, J = 5.8 Hz, 1H), 4.69 (br s, 1H), 4.10 (br s, 1H), 1.28 (d, J = 6.5 Hz, 6H). ^{13}C NMR (126 MHz, $CDCl_3$) δ 162.12, 159.56, 155.58, 150.15, 148.67, 140.88, 138.85, 129.48, 121.79, 120.44, 119.58, 117.51, 42.79, 22.85.

*N^2-([1,1′-Biphenyl]-4-yl)-N^4-isopropylpyrimidine-2,4-diamine (**6m**).* To a mixture of **3f** (0.12 mmol, 1.0 equiv) in EtOH (0.50 mL, 0.23 M) under a nitrogen atmosphere at rt were added 4-aminobiphenyl (0.18 mmol, 1.5 equiv) and a drop of 1 N HCl. The reaction mixture was heated to reflux and stirred at that temperature for 2 h; it was then cooled to rt. Purification using automated flash chromatography (MeOH/DCM) was followed by evaporation, giving **6m** as a white solid (0.024 g, 68%). TLC R_f 0.7 (10% MeOH/DCM). LC−MS (ESI) m/z: 305 $[M + H]^+$. ^1H NMR (400 MHz, chloroform-*d*) δ 8.05 (br s, 1H), 7.81 (br s, 1H), 7.73−7.63 (m, 2H), 7.63−7.48 (m, 4H), 7.48−7.37 (m, 2H), 7.34−7.28 (m, 1H), 5.93 (d, J = 6.2 Hz, 1H), 5.34 (br s, 1H), 4.07 (br s, 1H), 1.28 (d, J = 6.5 Hz, 6H). ^{13}C NMR (101 MHz, $CDCl_3$) δ 162.08, 140.77, 138.57, 135.42, 128.73, 127.41, 126.81, 126.72, 119.93, 43.25, 22.64.

*4-((4-(Isopropylamino)pyrimidin-2-yl)amino)benzamide (**6n**).* To a mixture of **3f** (0.10 mmol, 1.0 equiv) in EtOH (0.50 mL, 0.20 M) under a nitrogen atmosphere at rt were added 4-aminobenzamide (0.15 mmol, 1.5 equiv) and a drop of 1 N HCl. The reaction mixture was heated to reflux and stirred at that temperature for 2 h; it was then cooled to rt. Purification using automated flash chromatography (MeOH/DCM) was followed by evaporation, giving **6n** as a white solid (0.013 g, 48%). TLC R_f 0.3 (10% MeOH/DCM). LC−MS (ESI) m/z: 272 $[M + H]^+$. ^1H NMR (500 MHz, MeOD) δ 7.86−7.77 (m, 5H), 5.99 (d, J = 6.1 Hz, 1H), 4.23 (s, 1H), 1.27 (d, J = 6.5 Hz, 6H). ^{13}C NMR (126 MHz, MeOD) δ 170.82, 162.40, 159.20, 153.30, 144.38, 128.12, 125.46, 117.72, 97.87, 41.96, 21.31.

Biology. *Biochemical Assays.* Briefly, kinase reactions were conducted in triplicate, with a final assay volume of 10 μL, in black polystyrene 384-well plates (Corning #8849BC) using the Z′-LYTE Kinase Assay Kit (Tyr 2 peptide; Life Technologies). The reaction mixtures contained 500 μM ATP, 2 μM Tyr 2 peptide, and 6.25 or 12.50 nM recombinant human FLT3 (571−993 (SignalChem) in buffer [50 mM HEPES (pH 7.5), 10 mM $MgCl_2$, 1 mM EGTA, 0.01% Brij-35, 0.25 mM DTT]). A cocktail of FLT3, ATP, and peptide solution prepared in the above-described buffer (10 μL) was added to the plates using an automated plate filler (Wellmate, Matrix). DMSO stock solutions of the test compounds were then added in a dilution series by pin transfer (V&P Scientific) in nanoliter volumes. After 1 h of incubation at 23 °C, the reaction mixtures were quenched and processed according to the Z′-LYTE kit protocol and read on an automated EnVision (Perkin-Elmer) plate reader using a 400 nm excitation filter and a 535 nm emission filter. Fluorescence emission ratio data were processed using a custom script written in Pipeline Pilot (Accelrys) to transform data by $\log_{10}$ and calculate percent inhibition normalized in comparison to the means of positive and negative controls. Dose−response curves were plotted using GraphPad Prism software.

MV4-11 and BJ Cell Culture and Cytotoxicity Studies. BJ and MV4-11 cells were purchased from the American Type Culture Collection (ATCC, Manassas, Virginia). The cells were cultured in a humidified 5% CO_2 incubator at 37 °C according to vendor recommendations. BJ cells are cultured in EMEM media and MV4-11 cells are cultured in IMDM media purchased from ATCC; both lines are supplemented with 10% fetal bovine serum (GE Healthcare Life Sciences, Hyclone Laboratories, Logan, Utah). The cells were routinely tested for mycoplasma with the MycoAlert Mycoplasma Detection Kit (Lonza, Walkersville, MD).

Approximately 1000 BJ or 500 MV4-11 exponentially growing cells were plated per well (30 μL) in white polystyrene flat-bottom sterile 384-well tissue-culture-treated plates (Corning, Tewksbury, MA) and incubated overnight at 37 °C in a humidified 5% CO_2 incubator. Compound solutions (DMSO) were pin-transferred (V&P Scientific, San Diego, CA) the following day. Inhibition of proliferation was determined following 72 h of incubation using Promega Cell Titer Glo Reagent according to the manufacturer's recommendations.

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Luminescence was measured on an Envision plate reader (Perkin-Elmer).

MOLM13 Cell Culture. Parental MOLM13$^{FLT3\text{-}ITD}$ cells harboring FLT3-ITD (DSMZ, Brunswick, Germany) and MOLM13$^{FLT3\text{-}ITD/D835Y}$ cells carrying an additional D835Y mutation were maintained in RPMI 1640 medium supplemented with 10% fetal bovine serum (Life Technologies, Grand Island, NY) in a humidified 5% CO_2 incubator at 37 °C.[9,50] The MOLM13$^{FLT3\text{-}ITD/D835Y}$cells were maintained in the presence of 5μM tandutinib (LC Laboratories, Woburn, MA) and removed from the presence of the drug 3 days prior to experimentation

MOLM13 Cell Viability. Cell viability upon drug treatment was assessed using the MTT reagent (Roche Applied Science, Indianapolis, IN), as previously described.[50] Briefly, the cells were plated overnight in a humidified 5% CO_2 incubator at 37 °C. On day 2, the cells were treated with DMSO or drug reconstituted in DMSO for 72 h. The drug concentrations used were as follows: compound **1**, 1000−167 nM; compound **5e** 1600−6.55 nM; compound **6a**, 1000−6.55 nM; and compound **6k**, 1500−0.98 nM. Three individual experiments were conducted, with six replicates each. Cell viability was quantified as relative percentage to DMSO-treated cells, and cell viability curves were generated using GraphPad Prism software version 6.07.

Western Blot Analysis. Whole-cell lysates were made with radioimmunoprecipitation assay buffer (20 mM Tris−HCl (pH 7.5), 150 mM NaCl, 1 mM Na_2EDTA, 1 mM EGTA, 1% NP-40, 1% sodium deoxycholate, 0.1% SDS, and protease and phosphatase inhibitors). Immunoprecipitation was carried out with anti-FLT3 antibody and Protein A/G agarose beads from Santa Cruz Biotechnology (Dallas, TX). Invitrogen Bis−tris-gradient mini or midi gels were used for western blot analysis, followed by detection with enhanced chemiluminescence (ECL) reagent. Primary antibodies used were from Cell Signaling Technology (Danvers, MA): FLT3, phospho-FLT3, STAT5, and phospho-STAT5. Secondary antibodies were from Jackson Immunoresearch and Cell Signaling Technology.

In Vivo Pharmacokinetic Study. All animal studies were carried out under a St Jude Children's Research Hospital IACUC-approved protocol (#477). Female C57BL/6 mice, with an average weight of 18 g, were purchased from Charles River Laboratories (Wilmington, MA). Food and water were provided ad libitum. Twenty-four mice were divided into four dosage groups: 0, 3, 5, and 10 mg/kg. For each mouse, 0.1 mL of the compound suspension in formulation (5:50:45, EtOH/PEG400/PBS (pH 7.4), v/v/v) was administered by i.p. injection. Blood (0.1 mL) was collected retro-orbitally from a different mouse within each dosage group at 5, 15, and 30 min and 1, 4, and 24 h. The animals were euthanized via cardiac puncture after anesthesia at 48 h post injection. The blood samples were treated with 10 μL of EDTA sodium solution to prevent coagulation. The blood samples were kept under ice and centrifuged for 3 min at 13 200 rpm in a desktop centrifuge to collect plasma. The plasma samples (25 μL) were combined with 75 μL of internal standard (2 μM warfarin) in acetonitrile in a 96-well plate and centrifuged at 4000 rpm for 20 min at 4 °C. The supernatant (40 μL) was collected and mixed with two parts of deionized water and centrifuged again at 4000 rpm for 20 min at 4 °C. The plasma concentration was determined using an LC/MS−MS assay with multiple reaction monitoring detection (AB Sciex, Framingham, MA). The LC/MS−MS method is provided in the Supporting Information. The assay limit of quantification (LLOQ) was 13.7 nM in plasma.

The processed plasma concentration−time data were analyzed using noncompartmental analysis (NCA) in WinNonlin 6.0 with the Plasma (200−202) model type; all standard NCA parameters were estimated via default software settings, using predicted parameter estimates. If at least two-thirds of the observed concentrations were below the LLOQ, the mean concentration was treated as missing.

The AUC was calculated with the linear trapezoidal, linear interpolation rule using mean concentrations and nominal times. The terminal elimination rate (Lambda_z) and half-life (HL_Lambda_z) were determined using the default "Best Fit" method. The predicted AUC from the last time point to infinity (AUCINF_pred) was calculated as AUClast plus Clast(pred)/Lambda_z. CL was calculated as Dose/AUCINF_pred.

■ ASSOCIATED CONTENT

Supporting Information

The Supporting Information is available free of charge on the ACS Publications website at DOI: 10.1021/acsomega.7b00144.

Experimental procedures for ADME and MTD studies; additional experimental procedures for synthesis and characterization of compounds; additional synthetic schemes for preparation of intermediates; KinomeScan analysis for compound **1**; growth inhibition data against the BJ cell line for all compounds; FLT3 inhibition and MV4-11 cell proliferation data, including CI 95; solubility and permeability data for selected compounds; cellular permeability and PGP efflux assay data; in vivo blood chemistry data after i.p. dosing of compound **5e** in mice (PDF)

Experimental data for compounds **1**, **4**−**6**, **9**, **13**, **14**, **16**, **17**, and **19**−**22** (XLSX)

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The manuscript was prepared through contributions of all authors.

Notes

The authors declare no competing financial interest.

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■ ABBREVIATIONS

FLT3, FMS-like tyrosine kinase; SAR, structure−activity relationship; AML, acute myelogenous leukemia; FL, FLT3 ligand; ITD, internal tandem duplication; JM, juxtamembrane; wt, wild-type; DFG, Asp-Phe-Gly; SPR, structure−property relationship; CI, confidence interval; PGP, P-glycoprotein; TKD,

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tyrosine kinase domain; i.p, intraperitoneal; DCM, dichloromethane; EtOAc, ethyl acetate; EtOH, ethanol; MeOH, methanol

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