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Phospshoinositide 3-Kinase (PI3K)/Mammalian Target of Rapamycin (mTOR) Dual Inhibitors: Discovery and Structure−Activity Relationships of a Series of Quinoline and Quinoxaline Derivatives£

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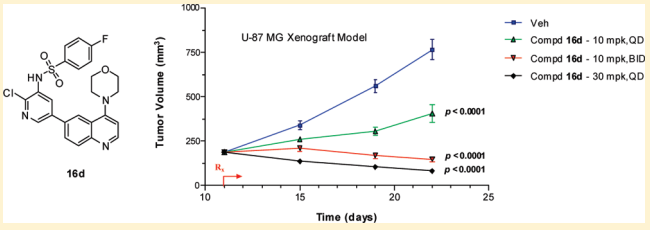
Supporting Information

ABSTRACT: The phosphoinositide 3-kinase (PI3K) family catalyzes the ATP-dependent phosphorylation of the 3′-hydroxyl group of phosphatidylinositols and plays an important role in cell growth and survival. There is abundant evidence demonstrating that PI3K signaling is dysregulated in many human cancers, suggesting that therapeutics targeting the PI3K pathway may have utility for the treatment of cancer. Our efforts to identify potent, efficacious, and orally available PI3K/mammalian target of rapamycin (mTOR) dual inhibitors resulted in the discovery of a series of substituted quinolines and quinoxalines derivatives. In this report, we describe the structure−activity relationships, selectivity, and pharmacokinetic data of this series and illustrate the in vivo pharmacodynamic and efficacy data for a representative compound.



INTRODUCTION

The phosphoinositide 3-kinase (PI3K) family catalyzes the ATP-dependent phosphorylation of the 3′-hydroxyl group of phosphatidylinositols. PI3Ks are subdivided into three classes, depending on sequence homology and substrate preferences. The class I PI3K family, comprising four isoforms (p110α, p110β, p110δ, and p110γ), generate phosphatidylinositol 3,4,5-triphosphate, (ptdIns (3,4,5)P3), a potent secondary messenger that triggers the activation of several downstream effectors, including the serine-threonine kinase, AKT (also known as protein kinase B or PKB). In response to PI3K activation, AKT is phosphorylated, which results in the triggering of a signal transduction cascade that ultimately stimulates mammalian target of rapamycin (mTOR) containing complex 1 (mTORC1). mTORC1 plays a key role in regulating cell growth, survival and proliferation by integrating diverse signaling inputs including growth factors, nutrient availability, and cellular energy levels.[1,2]

There is abundant evidence demonstrating that PI3K signaling is dysregulated in human cancers, with many tumors harboring somatic genetic alterations, that result in constitutive activation of the PI3K signaling network.[3,4] The most common genetic changes associated with constitutive PI3K signaling in cancer are loss of function mutations in the phosphatase and tensin homologue gene (PTEN), a lipid phosphatase that converts ptdIns (3,4,5)P3 to ptdIns (4,5)P2, and gain of function mutations in the PI3K catalytic subunit, p110α. These observations suggest that inhibitors targeting PI3K and mTOR may have utility as cancer therapeutics.[5]

In our efforts to inhibit the PI3K pathway, we identified an orally active dual PI3K/mTOR kinase inhibitor, *N*-(6-(6-chloro-5-(4-fluorophenylsulfonamido)pyridin-3-yl)benzo[*d*]thiazol-2-yl)-acetamide (**1**; Figure 1), which showed potent activities in both the biochemical (K_i values of 1, 2, 5, and 1 nM for PI3Kα, β, δ, γ, respectively, and an IC_{50} value of 2 nM for mTOR) and cellular (IC_{50} = 6.3 nM) assays.[6,7] In addition, compound **1** was active at low doses in vivo as determined by the inhibition of AKT (Ser 473) phosphorylation in a mouse liver pharmacodynamic assay (EC_{50} = 399 ng/mL) and by its ability to inhibit the growth of U-87 MG tumor xenografts (ED_{50} = 0.26 mg/kg). However, further investigations revealed that significant amounts of the deacetylated metabolite (2-aminobenzothiazole, **2**; Figure 1) were formed in both isolated hepatocytes and in vivo.[8] For example, when **1** was dosed orally in rats for 4 days at 0.3 mg/kg (q.d.), the ratio of *N*-acylamine **1** to amine **2** was ∼1.2 ($[AUC]_{0-24}$). The development of compound **1** was not desirable as the circulating

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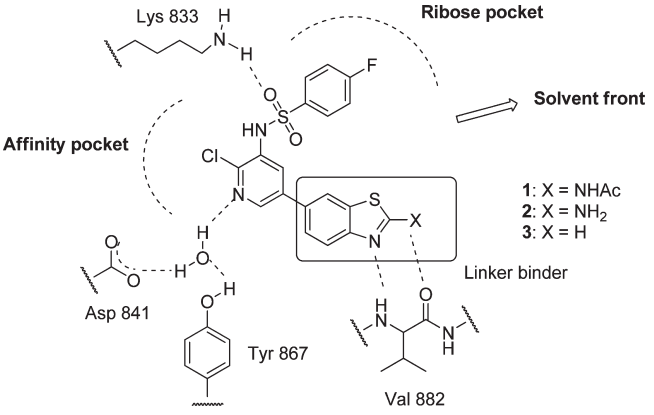

Figure 1. Key interactions of compounds **1**−**3** with PI3Kγ protein.

metabolite **2** is also a potent PI3K inhibitor (e.g., PI3Kα K_i = 5.6 nM), and the potential exists that reacetylation of **2** could occur in vivo. To circumvent this deacetylation problem, we investigated two different approaches. In the first approach, we sought to attenuate the in vivo stability of the *N*-acetyl group through the replacement of the benzothiazole ring with various 6,5-bicyclic heterocycles.[9] Those investigations led to the identification of the *N*-(imidazo[1,2-*b*]pyridazin-2-yl)acetamide series that proved to be stable toward deacetylation both in vitro and in vivo. In this investigation, we examined an alternative approach where we replaced the *N*-acetyl 2-aminobenzothiazole moiety in **1** with several 6,6-heterocyles that lack the *N*-acetyl moiety, therefore completely eliminating the potential of deacetylation.[10]

To aid in the design of novel inhibitors, we first analyzed the crystal structure of compound **1** bound to the active site of the PI3Kγ protein, which is highly homologous to the other class I PI3K isoforms.[11] The key interactions of compound **1** to the enzyme are illustrated in Figure 1. The *N*-acetyl benzothiazole group, also referred to as the linker binder moiety, forms two key hydrogen bonds with the backbone NH and carbonyl of Val 882. In this interaction, the thiazole nitrogen acts as the hydrogen bond acceptor and the NH of the *N*-acetyl group serves as the hydrogen bond donor. Other key interactions include a hydrogen bond to one of the sulfonamide oxygens from the Lys 833 side chain, and a hydrogen bond network between the nitrogen on the central pyridine ring and a water molecule that is bridged between the Asp 841 carboxylate and the Tyr 867 hydroxyl groups within the affinity pocket. Finally, the *para*-fluorophenyl group occupies the upper region of the ribose pocket.

Designing a linker binder without an *N*-acetamide group or a free exocyclic amine was crucial to resolving the deacetylation issue. To assess the binding contribution of the exocyclic amino group in **2**, compound **3** (Figure 2B) was prepared and its inhibitory activity against PI3Kα was determined to be K_i = 12.9 nM. The 10-fold decrease in activity was attributed to the difference in both the strength and orientation of the hydrogen bond interaction between the NH of **1** and CH of **3** with the carbonyl of Val 882 (Figure 2A vs B). When tested in the U-87 MG cellular assay, compound **3** showed an IC_{50} of more than 700 nM. The loss in cellular activity of **3** vs **1** (~200-fold) was more than the loss in enzyme activity (10-fold), suggesting that changes in the physicochemical properties of the compound led to reduced cell penetration or higher protein binding. We reasoned that by replacing the benzothiazole with a 6,6-heterocycle, such as a quinoline (Figure 2C), the key hydrogen bonding

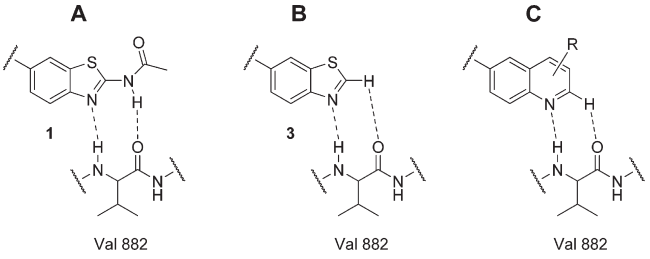

Figure 2. Interactions of the benzothiazole and quinoline linker binder groups with Val 882 in PI3Kα.

interaction to the NH of Val 882 would be maintained and the C(2)H would be closer to the Val 882 carbonyl.[12] Furthermore, the geometry of the C(2)H−carbonyl hydrogen bond appeared to be more optimal with the use of a 6,6-heterocyclic linker binder than with the benzothiazole group (Figure 2C vs B). More importantly, we postulated that it would be possible to modulate the physicochemical properties of such inhibitors by accessing the ribose pocket at either the 3- or 4-position of a 6,6-heterocyclic linker binder group thereby providing a means to improve both the enzyme and cellular potencies (Figure 2C; R-group). In this paper, we report the structure activity relationship (SAR) studies of the 6,6-heterocyclic linker binder systems that led to the identification of two new series of potent and selective PI3K/mTOR dual inhibitors with good pharmacokinetic and pharmacodynamic profiles. Initially, we examined several 6,6-heterocyclic systems as alternative linker binder groups and, subsequently, focused our SAR investigations on the 3-and 4-positions of quinoline and quinoxoline series. Optimizing the compounds in this way led to the discovery of quinoline **16d**, whose in vivo evaluation is described herein.

CHEMISTRY

Scheme 1 outlines the synthesis of benzothiazole **3** and compounds with various unsubstituted 6,6-heterocycles as shown in Table 1. The compounds were prepared through Suzuki coupling reactions[13] with either the bromide **4a** or the corresponding boronic acid pinacol ester **4b**, and either 5-bromobenzothiazole **5** or the appropriate 6,6- heterocycles. The requisite linker binder intermediates were either commercially available (**5**, **6a**, **6b**, **6e**, **6g**, and **6h**) or prepared as described below (**6c**, **6d**, and **6f**) in Scheme 2.

Scheme 2 shows the synthesis of the requisite heterocyclic intermediates **6c**, **6d**, and **6f**. The 2-chloro-1,5-naphthyridine intermediate **6c** was prepared from unsubstituted 1,5-naphthyridine (**8**) via N-oxide formation followed by chlorination with $POCl_3$ (Scheme 2A). This reaction gave a 1:1 mixture of the 4- and 2-chloro regioisomers **9** and **6c**, which were separated by silica gel chromatography. The synthesis of 1,7-naphthyridine derivative **6d** started by bromination of 2-cyano-3-methylpyridine (**10**) followed by a one carbon homologation using potassium cyanide to yield dicyanopyridine **11** (Scheme 2B). The key cyclization was performed in acetic acid in the presence of HBr to yield the 8-bromo-1,7-naphthyridin-6-amine **12**. The bromine of **12** was removed via catalytic hydrogenation and the amino group was converted directly to a triflate by diazotization of the amine followed by displacement with triflic acid to afford **6d**. The pinacol ester of quinoxaline **6f** was prepared from the corresponding bromide (**13**) by treatment with bis(pinacolato)-diborane (Scheme 2C).

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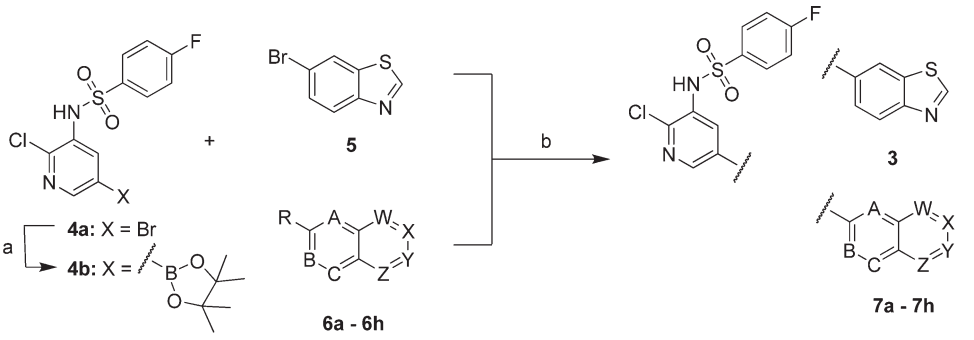
Scheme 1. Synthesis of Compounds in Table 1[a]



6 and 7	A	B	C	W	X	Y	Z	R in 6
A	CH	CH	CH	CH	CH	CH	**N**	$B(OH)_2$
B	CH	CH	CH	CH	CH	**N**	CH	Br
C	**N**	CH	CH	CH	CH	CH	**N**	Cl
D	CH	**N**	CH	CH	CH	CH	**N**	OTf
E	CH	CH	**N**	CH	CH	CH	**N**	Br
F	CH	CH	CH	**N**	CH	CH	**N**	$B(-OC(Me)_2C(Me)_2O-)$
G	CH	CH	CH	CH	**N**	CH	**N**	Br
H	CH	CH	CH	CH	CH	**N**	**N**	Br

[a] Reagents and conditions: (a) $PdCl_2$dppf, bis(pinacolato)diborane, potassium acetate, 1,4-dioxane, 120 °C; (b) $PdCl_2$dppf or $Pd(PPh_3)_4$, potassium or sodium carbonate, 1,4-dioxane, water, 90−100 °C.

Scheme 3 shows the synthesis of 4-substituted quinolines that are illustrated in Table 2. In general, the targeted compounds **16a**−**f** were prepared by Suzuki reactions of **4a** or **4b** with amino-substituted quinolines **15b**−**15e** that were derived from **14** via S_N2' reactions. Alternatively, the chloro intermediate **16a** was treated with a variety of secondary amines to give derivatives **16f**−**16i** or with aryl boronic acids or esters to provide the corresponding aromatic derivatives **16j**−**16m**. In the case of compound **16e**, the requisite Suzuki coupling partner was boronic acid **15e**. This intermediate was synthesized by first converting **14** to the corresponding boronic ester (**15a**) and then substituting the 4-chloro with 1-(pyridine-4-ylmethyl)piperidine. The boronic ester was hydrolyzed to the boronic acid during the substitution reaction to produce **15e**.

Scheme 4 outlines the synthesis of 3-substituted quinoxalines (**18a**−**18s**). In the majority of cases, the compounds were prepared from the intermediates **18a** or **18b** that were obtained by the coupling of boronic ester **4b** with either 7-bromo-2-chloroquinoxaline (**17a**)[14] or the more reactive 7-bromo-2-fluoroquinoxaline (**17b**). The latter compound, **17b**, was obtained by treating **17a** with tetra-*n*-butylammonium fluoride (TBAF) in DMSO.[15] The final compounds were prepared either via S_N2' reactions of fluoroquinoxaline **18b** with the appropriate amines to give **18f**−**r** or via a Suzuki reaction of **18b** with pyridin-4-yl boronic acid to provide **18e**. Compounds **18c** and **18d** were prepared by coupling **4b** with **17c** and **17d**, which were derived from 7-bromo-2-fluoroquinoxaline (**17b**). Deprotection of the Boc-protected piperidine derivative **18r** with trifluoroacetic acid (TFA) provided the corresponding piperidine **18s**.

RESULTS AND DISCUSSION

In general, the compounds described in this paper were inhibitors of PI3Kα, β, δ, γ, and mTOR; however, for simplicity, the following SAR discussion will focus on inhibition of PI3Kα as well as activities in the U-87 MG cellular assay that measures the inhibition of pAKT (Ser 473). Selectivity profiles of the most potent derivatives are reported in Table 4 and in the Supporting Information.

To test the hypothesis that 6,6-bicyclic heterocycles could serve as replacements for the benzothiazole linker binder, quinoline, isoquinoline, quinoxaline, quinazoline, cinnoline, and naphthyridine analogues of compound **1** were prepared and evaluated. The results are shown in Table 1. Quinoline **7a** was ~2-fold more potent in both enzyme and cellular assays than benzothiazole **3** and achieved a similar enzyme potency as aminobenzothiazole **2**, thereby illustrating the ability of a 6,6-heterocycle to serve as an excellent linker binder moiety. However, moving the nitrogen to the 2-position, as in isoquinoline **7b**, resulted in dramatic loss of potency (K_i = 375 nM), indicating that the position of the hydrogen bond acceptor was extremely important. For the remainder of the derivatives shown in Table 1, we maintained the quinoline nitrogen of **7a** and systematically added one additional nitrogen at each of the other six positions of the bicyclic ring. When the additional nitrogen was incorporated at the 3, 4, or 5 positions of the quinoline ring system, similar levels of enzyme activities were obtained (i.e., quinazoline **7g**, quinoxaline **7f**, and 1,5-naphthyridine **7c**). On the other hand, adding the additional nitrogen to the 7 or 8 positions gave two isomeric naphthyridines (**7d** and **7e**) that exhibited >100-fold drop in enzyme potency compared to **7a**. These results demonstrated that having an additional hydrogen bond acceptor at the same side as N1-nitrogen was detrimental to the molecule's ability to bind to the enzyme. For example, the N-8 nitrogen of **7e** would have an unfavorable interaction with the backbone carbonyl of Glu 880. In addition, the aromatic CH at the 8 position of **7a** also can form a favorable interaction to the carbonyl of Glu 880, which is lacking in **7e**. In the case of **7d**,

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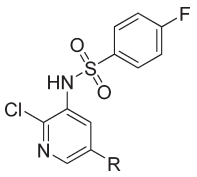
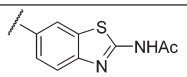
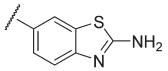
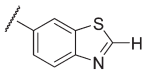
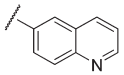
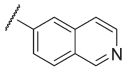
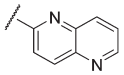
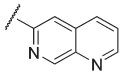
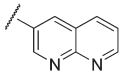
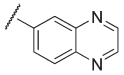
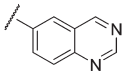
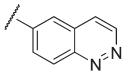
Table 1. Enzyme and Cellular Assay Results of Various 6,6-Bicyclic Heterocyclic Derivatives[a]



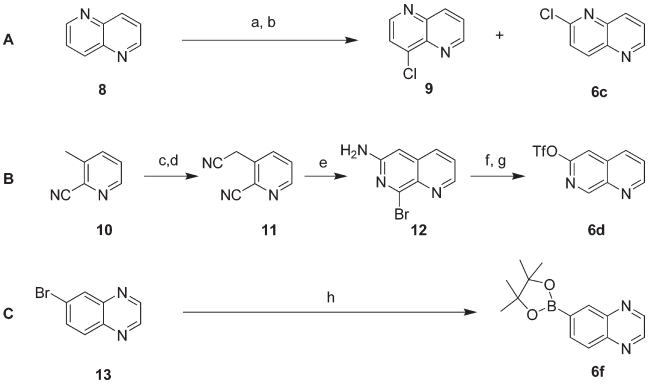
Compd Number	R	PI3Kα Ki (nM)[b]	U-87 MG IC_{50} (nM)[b]
1		1.2 ± 0.9	6.3 ± 6.5
2		5.6 ± 3.0	44
3		12.9 ± 8.2	769 ± 336
7a		2.1 ± 0.4	377
7b		375 ± 32	>10,000
7c		5.1 ± 0.4	784
7d		200 ± 36	>10,000
7e		856 ± 38	NA
7f		3.1 ± 0.3	1450
7g		2.6 ± 0.8	23
7h		38 ± 0.07	1920

[a] The inhibition of PI3Kα activity was determined using a modified in vitro AlphaScreen assay with human p110α enzyme. Cellular IC_{50} values were determined by AKT(Ser 473) phosphorylation assay using U-87 MG cells. [b] The results are reported as average ± SD. The results without SD are from a single measurement.

the nitrogen at the 7 position is adjacent to the face of Tyr 867, which is also a less favorable interaction than the CH of **7a**. Similarly cinnoline **7h**, which lacks the CH as hydrogen bond donor to Val 882, also showed a 15-fold drop in potency. It is likely that the nitrogen at the 2-position of the cinnoline core causes an unfavorable interaction with the carbonyl of Val 882 and therefore leads to diminished affinity.

Having identified four 6,6-heterocyclic systems that showed K_i values of <10 nM (compounds **7a**, **7c**, **7f**, and **7g**), we focused on the quinoline and quinoxaline scaffolds (**7a** and **7f**) for further

Scheme 2. Synthesis of Intermediates 6c, 6d, and 6f[a]



[a] Reagents and conditions: (a) 3-chloroperoxybenzoic acid, DCM, room temperature; (b) $POCl_3$, 100 °C; (c) NBS, CCl_4, reflux; (d) KCN, MeOH, room temperature; (e) HBr, AcOH, 0 °C; (f) Pd/C, KOH, H_2, room temperature; (g) $NaNO_2$, trifluoromethanesulfonic acid, DMF, room temperature; (h) $PdCl_2$dppf, bis(pinacolato)diborane, potassium acetate, 1,4-dioxane, 120 °C.

optimization based on the following considerations: (1) We found that quinazoline **7g** was unstable in weakly acidic conditions such as 0.1% TFA in water. It is likely that the quinazoline ring undergoes hydrate formation, a phenomenon well-known for this type of system.[16,17] (2) Modifications at either the 3- or 4-position of the 6,6-bicyclic systems were more readily accessible from the quinoline and the quinoxaline derivative (**7a** and **7f**) relative to 1,5-naphthyridine analogue (**7c**). (3) Furthermore, the additional nitrogen at the 5-position of **7c** seemed to offer no significant advantage over quinoline **7a**.

To improve the cellular potency of compound **7a**, a variety of 4-substituted quinolines were synthesized and evaluated. The results of the PI3Kα enzyme and U-87 MG cellular assays are shown in Table 2. The methoxyethylamine derivative **16c** showed a 35-fold decrease in the enzyme potency relative to **7a**. We postulated this loss of potency was due to the presence of a tautomerizable NH proton on the substituent that would diminish the ability of the quinoline nitrogen to serve as a hydrogen bond acceptor. On the other hand, tertiary amines at the 4-position, such as the dimethyl amino analogue **16b**, were well tolerated. Even bulkier groups were tolerated in the ribose pocket as shown by the single-digit nanomolar enzyme activity from piperidine **16f**. More importantly, introducing polar functionalities on the piperidine ring proved to be fruitful in improving the cellular potency. For example, the 4-hydroxylpiperidine analogue **16g** was 3-fold more potent in the cellular assay than **16f** and the morpholine derivative **16d** was 30-fold more potent, with an IC_{50} of 16 nM. Compound **16d** represented the first example of a modified linker binder derivative showing an IC_{50} < 20 nM in the cellular assay. The piperazine analogues **16h** and **16e** also showed cellular potencies comparable to that of **16d**, indicating that polar groups were preferred in this region. As lipophilicity increased, as was the case for the *N*-benzyl piperazine analogue **16i**, so did the enzyme-to-cell shift. This trend also held true for aromatic substituents; as the lipophilicity of the substituents decreased in going from phenyl (**16j**) to 4-dimethylaminophenyl (**16m**) to 3-pyridyl (**16k**) and 4-pyridyl (**16l**), incremental improvements in cellular potencies (127 to 4 nM) were observed.

With the success in the 4-substituted quinoline series, we turned our attention to 3-substituted quinoxalines to probe the

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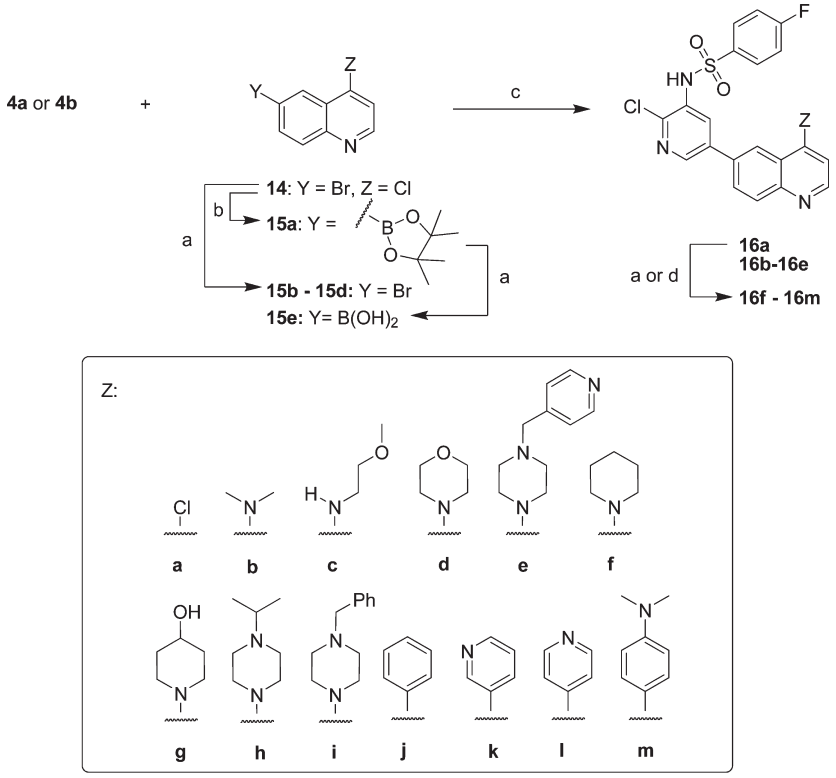
Scheme 3. Synthesis of Compounds with 4-Substituted Quinolines[a]



[a] Reagents and conditions: (a) amine, DMF or DMSO, 90−120 °C; (b) $PdCl_2$dppf, bis(pinacolato)diborane, potassium acetate, 1,4-dioxane, 120 °C; (c) $PdCl_2$dppf, potassium or sodium carbonate, 1,4-dioxane, water, 90−100 °C; (d) For **16j**: phenylboronic acid, FibreCat, sodium carbonate, 1,4-dioxane, water, 120 °C; for **16k**−**16m**: aryl boronic acid or aryl boronic ester, dichlorobis(*t*-butylphenylphosphine)palladium, potassium acetate, *n*-butanol, water, 100 °C.

structure activity relationships around a different area of the ribose pocket. The results are shown in Table 3. When the 3 position of quinoxaline **7f** was substituted by a methoxyethylamine (**18f**), the enzymatic activity was increased in contrast to the observed decrease in the corresponding quinoline analogue **16c**. This is likely due to the fact that the 3-methoxyethylamine NH proton in **18f** cannot tautomerize with the N-1 nitrogen that is involved in the crucial hydrogen bonding interaction with Val 882, unlike the corresponding quinoline analogue **16c**. As a result, there was little change in potency in going from a secondary amine to a tertiary amine (**18f** vs **18g**) in this series. In addition, both **18f** and **18g** showed more than a 25-fold improvement in the cellular potencies compared to the parent quinoxaline **7f**. Similar to the quinoline series, bulkier groups were tolerated in the quinoxaline series as well, as seen by analogues **18h**, **18i**, and **18j**. However, these analogues also showed relatively large enzyme-to-cell shifts, which may be attributed to the compounds' overall increased lipophilicity. Introducing polar functionalities in this area improved cellular potency, as shown by analogues **18k** and **18l**. Surprisingly, the 3-piperidine derivative **18s** showed a significant loss in cellular activity compared to **18h**.

While dimethylamine at the 3-position in the quinoxaline core was well tolerated (**18c**; U-87 MG cellular IC_{50} = 44 nM), larger cell shifts with highly lipophilic substituents tended to be associated with compounds bearing cyclic amines, such as **18m**, **18n**, and **18o**. When heteroatoms were introduced to these groups, the cellular potencies were improved, as shown with analogues **18p**, **18q**, and **18d**. The 4-pyridyl derivative **18e** also showed good activity in the cellular assay. In general, the cellular potency in the 3-substituted quinoxaline series was less sensitive to substitution than in the case of the 4-substituted quinoline series. This observation may be rationalized in that substituents in the 3-position of the quinoxalines project away from the binding pocket and more toward the solvent front than do groups on the 4-position of the quinolines.

To examine this in more detail, we obtained an X-ray crystal structure of quinoline **16d** bound to PI3Kγ protein (Figure 3A). The binding mode of **16d** was similar to the benzothiazole compound **1**. As was observed in the crystal structure of benzothiazole compound **1** (illustrated in Figure 1), the quinoline nitrogen forms a hydrogen bond with the Val 882 backbone NH and a water molecule is bridged between the Asp 841 carboxylate, the Tyr 867 hydroxyl group, and the nitrogen on the center pyridine ring. The morpholine ring in **16d** occupies the ribose pocket (the space above the quinoline ring) that was unoccupied in the crystal structure of compound **1**. The quinoxaline compound **18d** was modeled into the X-ray structure of the enzyme (Figure 3B). The space-filling model on the right shows that the morpholine ring extends toward the solvent front rather than into the ribose pocket. This is consistent with the observation that the cellular potency in the 3 substituted quinoxalines series was less sensitive to substitution than the corresponding quinoline analogues.

The SAR investigations described above enabled the identification of several potent PI3K/mTOR dual inhibitors with IC_{50}

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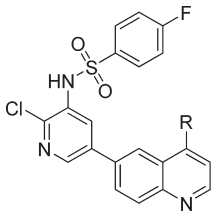
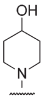
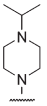
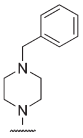
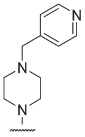
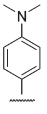
Table 2. Enzyme and Cellular Assay Results of 4-Substituted Quinoline Derivatives.[a]



Compd Number	R	PI3Kα Ki (nM)[b]	U-87 MG IC_{50} (nM)[b]
7a		2.1 ± 0.4	377
16c		69 ± 13	1020
16b		5.7 ± 0.8	223
16f		1.9 ± 0.3	498
16g		0.9 ± 0.07	150
16d		0.6 ± 0.5	16 ± 5.8
16h		1.8 ± 0.1	28
16i		1.0 ± 0.2	136
16e		0.9 ± 0.003	18
16j		1.3 ± 0.4	127
16k		0.7 ± 0.02	12 ± 1.7
16l		0.8 ± 0.07	3.7 ± 0.7
16m		0.7 ± 0.005	61

[a] The inhibition of PI3Kα activity was determined using a modified in vitro AlphaScreen assay with human p110α enzyme. Cellular IC_{50} values were determined by AKT(Ser 473) phosphorylation assay using U-87 MG cells. [b] The results are reported as average ± SD. The results without SD are from a single measurement.

values of ≤20 nM in the U-87 MG cellular assay (**16d**, **16e**, **16k**, **16l**, **18d**, **18k**, and **18q**).[18] The in vitro profiles of these derivatives are reported in Table 4. All compounds were potent against all of the class I PI3K isoforms, mTOR, and hVPS34, a class III PI3K. The compounds were also potent inhibitors of the protein kinase DNA-PK (DNA-dependent serine/threonine protein kinase), which contains a catalytic domain homologous to the PI3Ks, but were selective against other protein kinases.[19]

The pharmacokinetic properties of these compounds were also examined and the results are shown in Table 5. All but one compound, **16e**, showed good stability in rat and human liver microsomes, and therefore, they were evaluated in rat in vivo pharmacokinetic studies. In general, all compounds tested showed low in vivo clearance (0.02−0.17 L/h/kg; 0.6−1% of rat liver blood flow rate), low to moderate volume of distribution (0.2−1.2 L/kg), and a moderate to long mean residence time (MRT; 2−15 h). When dosed orally, all except compound **18k**, demonstrated excellent bioavailability (%F = 70−92) and high exposures (AUC).

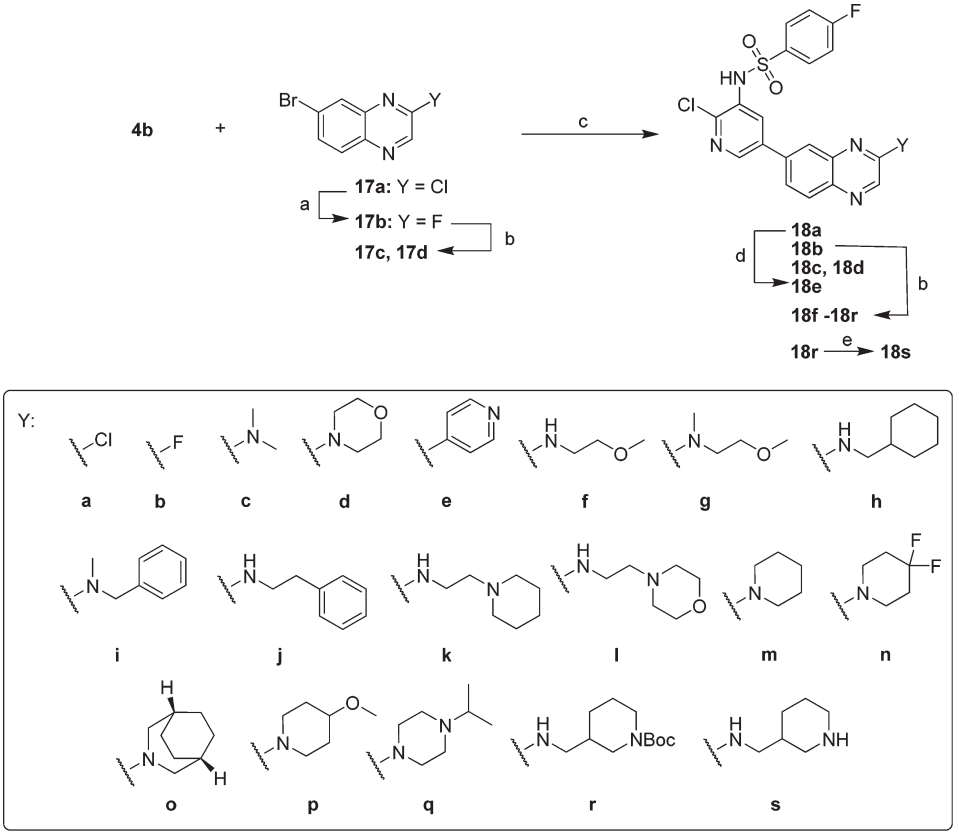
Compounds with acceptable pharmokinetic properties (**16d**, **16k**, **16l**, **18d**, and **18q**) were tested in a mouse liver pharmacodynamic (PD) model in which the inhibition of HGF induced AKT (Ser 473) phosphorylation was measured.[20] All compounds were dosed orally at 0.3, 1, and 3 mg/kg, and after 3 or 4 h, the mice were injected with 12 μg of hepatocyte growth factor (HGF) to activate PI3K-dependent AKT phosphorylation in the liver. The levels of AKT (Ser 473) phosphorylation were determined five minutes post HGF administration by a quantitative electrochemiluminescence immunoassay. The results are shown in Table 6. All five compounds showed dose-dependent inhibition of AKT (Ser 473) phosphorylation, indicating that they effectively inhibited PI3K in vivo. The dose−response data for a representative example, quinoline **16d**, is shown graphically in Figure 4A. In addition to testing the compounds at the 3 or 4 h time points, we were interested in evaluating the duration of the compound's inhibitory effect in vivo. The time-course PD for quinoline **16d** is illustrated in Figure 4B. In mice dosed orally at 3 mg/kg, compound **16d** inhibited the phosphorylation of AKT (Ser 473) at 96% after 1 h and 72% after 6 h, and the effect was significantly diminished after 24 h (29% inhibition).

The promising pharmacodynamic results of compound **16d** in the mouse liver model prompted us to evaluate it in a PTEN-null U-87 MG glioblastoma xenograft model.[21] Compound **16d** was dosed orally at 10 mg/kg daily, 30 mg/kg daily, or at 10 mg/kg twice daily for 12 days (Figure 5).[22] At 10 mg/kg daily dosing, the tumor growth was suppressed 62% compared to the vehicle group. Furthermore, tumor regression was observed when dosed either at 10 mg/kg bid or 30 mg/kg qd, showing that compound **16d** was efficacious in this model.

CONCLUSION

We have identified two series of potent PI3K/mTOR dual inhibitors with excellent pharmacokinetic properties and in vivo efficacies. These compounds were designed to avoid the issue of deacetylation encountered with the early lead compound **1**. By adopting 6,6-heterocycles as linker binders, we were able to eliminate the acetyl amino hydrogen bonding donor without sacrificing the enzyme potency. Furthermore, we demonstrated that by incorporating suitable substituents either at the 4-position of the quinoline or the 3-position of the quinoxaline rings, excellent cellular potencies could also be achieved. In addition,

Scheme 4. Synthesis of 3-Substituted Quinoxalines[a]



[a] Reagents and conditions: (a) TBAF, DMSO, room temperature; (b) amine, DMSO, or DMF, 60−100 °C; (c) $PdCl_2$dppf or dichlorobis(di-*t*-butylphenylphosphine)palladium, potassium carbonate, 1,4-dioxane, water, 90−100 °C; (d) pyridin-4-yl boronic acid, dichlorobis(di-*t*-butylphenylphosphine)palladium, potassium acetate, 1,4-dioxane, water, 90 °C; (e) TFA, DCM, room temperature.

these results supported the hypothesis that the ribose pocket of the enzyme can be effectively utilized in optimizing both the potency and the physicochemical properties of PI3K inhibitors. Furthermore, we demonstrated that a representative compound, **16d**, successfully blocked the targeted PI3K pathway in a mouse PD model and inhibited tumor growth in a U-87 MG xenograft model.

EXPERIMENTAL SECTION

Chemistry. *General.* Unless otherwise noted, all materials were obtained from commercial suppliers and used without further purification. Anhydrous solvents were obtained from Aldrich, Acros, or EM Science and used directly. All reactions involving air- or moisture-sensitive reagents were performed under a nitrogen or argon atmosphere. Microwave-assisted reactions were conducted with either an initiator from Biotage, Uppsala, Sweden, or Explorer from CEM, Matthews, North Calorina. Silica gel chromatography was performed using either glass columns packed with silica gel (200−400 mesh, Aldrich Chemical) or prepacked silica gel cartridges (Biotage or Redisep). Preparative HPLC was performed with Varian Prostar using method: [A] Phenomenex Synergi, MAX-RP column (150 × 50 mm, 10 μ) with a 80 mL/min flow rate using a gradient of 5−100% [0.1% TFA in acetonitrile] in [0.1% TFA in water] over 20 min; [B] Phenomenex Gemni, C18 column (100 × 30 mm, 5 μ) with a 40 mL/min flow rate using a gradient of 5−100% [0.1% TFA in acetonitrile] in [0.1% TFA in water] over 20 min. NMR spectra were determined with a Bruker 300 MHz or DRX 400 MHz spectrometer. Chemical shifts were reported in parts per million (ppm, δ units). All final compounds were purified to >95% purity as determined by LC/MS obtained on an Agilent 1100 spectrometer using the following methods: [A] Agilent SB-C18 column (50 × 3.0 mm, 2.5 μ) at 40 °C with a 1.5 mL/min flow rate using a gradient of 5−95% [0.1% TFA in acetonitrile] in [0.1% TFA in water] over 3.5 min; [B] Phenomenex Gemni NX C18 column (50 × 3.0 mm, 3 μ) at 40 °C with a 1.5 mL/min flow rate using a gradient of 5−95% [0.1% formic acid in acetonitrile] in [0.1% formic acid in water] over 3.5 min. Low-resolution mass spectral (MS) data were obtained at the same time of the purity determination on the LC/MS instrument using ES ionization mode (positive). High-resolution exact mass measurements were performed using electrospray ionization (positive mode) on a quadrupole time-of-flight (QTOF) mass spectrometer (Xevo Q-TOF, Waters Inc.). The instrument was calibrated immediately before performing the exact mass measurements using sodium formate cluster ions; the resulting mass accuracy for all calibrant ions was better than 1.5 ppm. DMSO stock solutions (10 mM) were diluted 2000 times with 50/50 acetonitrile/water. Each sample (2 μL) was introduced to the mass spectrometer by flow injection using 1:1 water/acetonitrile as the carrier solvent with a flow rate of 100 μL/min. MS data were acquired from 100 to 1000 m/z with an acquisition rate of 1.0 scan/s. Reserpine (~40 fmol/μL) was used as a lock mass reference and was ionized using a second orthogonal sprayer.

General Boronic Ester Formation Method. To an appropriately sized reaction vessel was added a halide (1 equiv), bis(pinacolato)diborane (1.1 equiv), potassium acetate (2.0 equiv), and $PdCl_2$dppf (0.05 equiv) in 1,4-dioxane. The mixture was deoxygenated by bubbling nitrogen through it for 5 min. The reaction mixture was heated at 120 °C by

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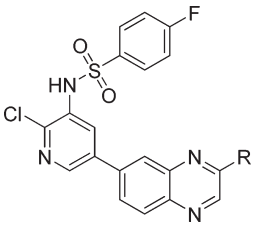
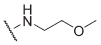
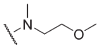
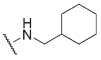
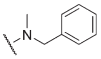
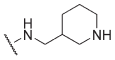
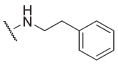
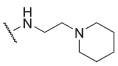
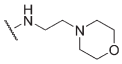
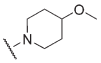
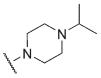
Table 3. Enzyme and Cellular Assay Results of 3-Substituted Quinoxaline Derivatives[a]



Compd Number	R	PI3Kα Ki (nM)[b]	U-87 MG IC_{50} (nM)[b]
7f		3.1 ± 0.3	1450
18f		0.9 ± 0.01	54
18g		1.0 ± 0.2	56
18h		0.8 ± 0.09	281
18i		1.4 ± 0.01	258
18s		3.3 ± 0.5	2710
18j		0.8 ± 0.008	241
18k		3.1 ± 0.2	18
18l		1.2 ± 0.009	44
18c		1.7 ± 0.6	44
18m		0.9 ± 0.06	128
18n		0.6 ± 0.05	189
18o		1.0 ± 0 .07	247
18p		1.4 ± 0.3	46
18q		0.8 ± 0.03	17
18d		0.8 ± 0.1	20±14
18e		1.0 ± 0.4	24

[a] The inhibition of PI3Kα activity was determined using a modified in vitro AlphaScreen assay with human p110α enzyme. Cellular IC_{50} values were determined by AKT(Ser 473) phosphorylation assay using U-87 MG cells. [b] The results are reported as average ± SD. The results without SD are from a single measurement.

conventional heating or irradiated with microwave at 120 °C, until the starting halide was consumed. The reaction mixture was allowed to cool to room temperature then partitioned between water and EtOAc. The aqueous phase was extracted with EtOAc. The combined organic phases were washed with saturated aqueous NaCl. The organic phase was dried over sodium sulfate, filtered, and concentrated in vacuo. If necessary, the crude product was purified by silica gel column chromatography.

General Suzuki Coupling Method. To an appropriately sized reaction vessel was added a halide (1 equiv), a boronic ester or acid (1.1 equiv), potassium or sodium carbonate (2 M aqueous solution, 3 equiv), palladium catalyst ($PdCl_2dppf$, $Pd(PPh_3)_4$, or FibreCat, 0.05 to 0.1 equiv) in 1, 4-dioxane. The mixture was deoxygenated by bubbling nitrogen through it for 5 min. The mixture was heated at 90 °C by conventional heating or irradiated with microwave at 100 °C, until the starting halide was consumed. The reaction mixture was allowed to cool to room temperature and was partitioned between water and EtOAc. The aqueous phase was extracted with EtOAc. The combined organic phases were washed with saturated aqueous NaCl. The organic phase was dried over sodium sulfate, filtered, and concentrated in vacuo. If necessary, the crude product was purified by silica gel column chromatography.

General S_N2' Method on Aryl Halides. To a solution of aryl halide (1 equiv) in DMF or DMSO was added an amine (2−10 equiv). The reaction mixture was heated to 60−100 °C until the starting aryl halide was consumed. The reaction mixture was allowed to cool to room temperature then directly subjected to chromatographic purification or partitioned between water and organic solvent, extracted with more organic solvent. The combined organic phases were dried, filtered, and concentrated, then purified by silica gel column chromatography.

*N-(5-(Benzo[d]thiazol-6-yl)-2-chloropyridin-3-yl)-4-fluorobenzenesulfonamide (**3**).* This compound was prepared from **4b** (0.212 g, 0.514 mmol) and 6-bromobenzo[d]thiazole (0.100 g, 0.467 mmol) according to the general Suzuki coupling method to obtain the title compound (0.150 g, 77%) as a tan solid. MS (ESI pos. ion) *m/z*: calc'd for $C_{18}H_{11}ClFN_3O_2S_2$: 419.0; found 419.9 (M+1). HRMS calc'd for $C_{18}H_{11}ClFN_3O_2S_2$ (M + H): 420.0037; found 420.0045. ^{1}H NMR (400 MHz, DMSO-d_6) δ 7.43 (t, *J* = 8.71 Hz, 2 H), 7.77−7.91 (m, 3 H), 8.08 (d, *J* = 2.15 Hz, 1 H), 8.22 (d, *J* = 8.41 Hz, 1 H), 8.55 (d, *J* = 1.37 Hz, 1 H), 8.63 (d, *J* = 1.96 Hz, 1 H), 9.48 (s, 1 H), 10.54 (br s, 1 H).

*N-(5-Bromo-2-chloropyridin-3-yl)-4-fluorobenzenesulfonamide (**4a**). (a). N-(5-bromo-2-chloro-3-pyridinyl)-4-fluoro-N-((4-fluorophenyl)sulfonyl)benzenesulfonamide.* To a solution of 5-bromo-2-chloropyridin-3-amine (20.0 g, 96.4 mmol) in pyridine (150 mL) was added 4-fluorobenzene-1-sulfonyl chloride (41.3 g, 212 mmol). The resulting mixture was heated to 100 °C under N_2. The reaction was allowed to cool to room temperature. Water (100 mL) was added and the mixture was stirred at room temperature for 1 h. The resulting precipitate was collected by filtration, washed with water, and air-dried to afford the title compound (43.0 g, 85%) as an off-white solid. MS (ESI pos. ion) *m/z*: calc'd for $C_{17}H_{10}BrClF_2N_2O_4S_2$: 521.9; found 522.7, 524.7 (M+1, M+3). ^{1}H NMR (300 MHz, $CDCl_3$) δ 7.23−7.35 (m, 4 H), 7.61 (d, *J* = 2.19 Hz, 1 H), 7.95−8.06 (m, 4 H), 8.55 (d, *J* = 2.34 Hz, 1 H).

*(b). N-(5-Bromo-2-chloropyridin-3-yl)-4-fluorobenzenesulfonamide (**4a**).* To a suspension of *N*-(5-bromo-2-chloro-3-pyridinyl)-4-fluoro-*N*-((4-fluorophenyl)sulfonyl)benzenesulfonamide (36.6 g, 69.9 mmol) in MeOH (400 mL) was added potassium carbonate (21.2 g, 154 mmol) and water (5 mL). The resulting mixture was stirred at room temperature for 20 h. The solvent was removed in vacuo. The residue was suspended in water and the pH was adjusted to 6 using 5 N HCl. The aqueous phase was extracted with EtOAc (4 × 200 mL). The combined organic phases were dried over magnesium sulfate, filtered, and concentrated. Crystallization from 30% acetone in hexanes afforded the title compound (21.5 g) as an off-white solid. The mother liquor was concentrated and purified by silica gel chromatography (20% acetone in hexanes) to afford another batch of the title compound (2.7 g) as a tan

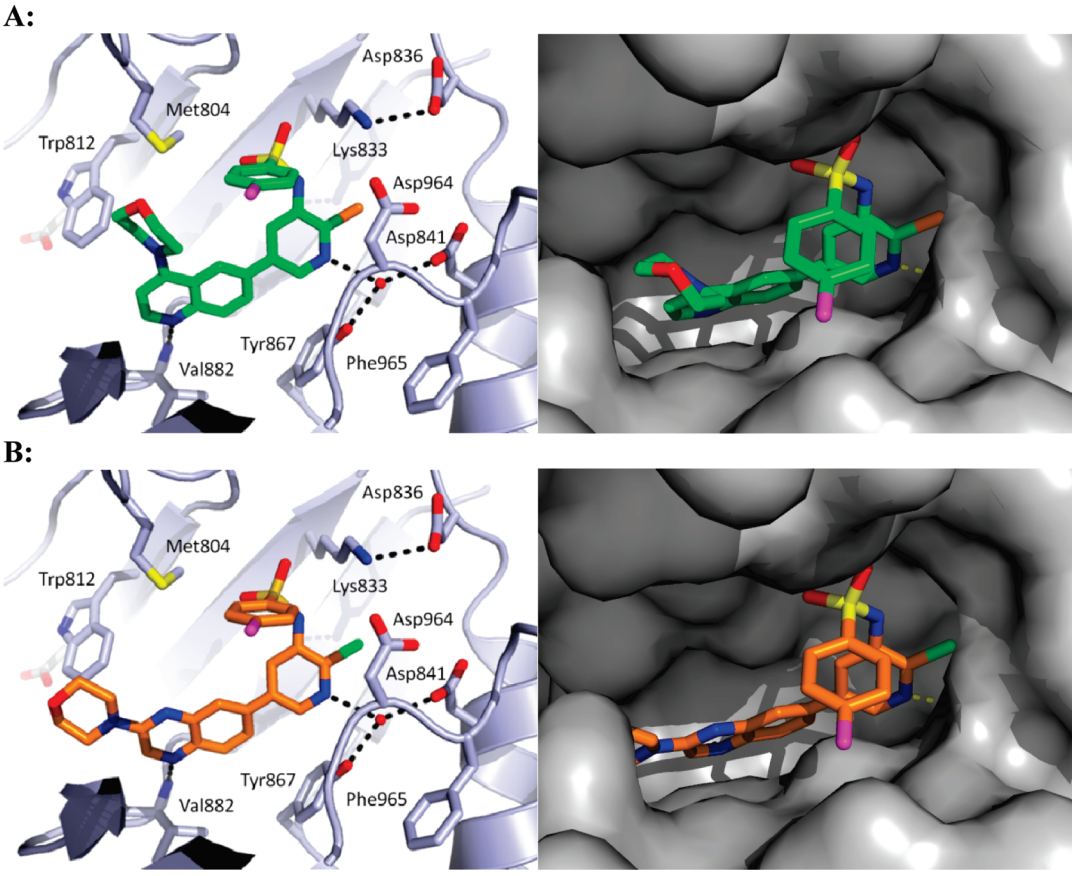


Figure 3. (A) X-ray crystal structure of **16d** (green) in PI3Kγ. (B) Modeled structure of **18d** (orange) in PI3Kγ.

Table 4. In Vitro Profiles of Selected Compounds[a]

	PI3K IC_{50} (nM)						
compd number	α	β	γ	δ	mTOR IC_{50} (nM)	hVPS34 IC_{50} (nM)	DNA-PK IC_{50} (nM)
16d	4.6 ± 3	13 ± 10	8.1 ± 3	4.3 ± 2	3.9 ± 1	11 ± 1	2.3 ± 0.5
16e	4.6 ± 2	15 ± 0.7	6.5 ± 1	6.0 ± 0.1	3.9 ± 2	–	–
16k	5.4 ± 3	10 ± 7	7.7 ± 3	5.9 ± 3	1.6 ± 2	2.9 ± 0.08	–
16l	6.5 ± 4	15 ± 0.3	13 ± 1	6.0 ± 0.4	0.41 ± 0.2	5.3 ± 0.05	–
18d	3.1 ± 0.2	15 ± 12	2.4 ± 0.3	4.4 ± 0.4	5.0 ± 1	5.0 ± 2	3.4 ± 1
18l	12 ± 0.8	9.8 ± 6	16 ± 1.	6.0 ± 0.5	5.3 ± 0.5	–	–
18q	5.4 ± 1	9.6 ± 4	13 ± 2	5.6 ± 0.5	37 ± 19	–	–

[a] The results are reported as average ± SD. The results without SD are from single measurement. Details of the assays are reported in the Supporting Information.

solid. The total combined yield was 95%. MS (ESI pos. ion) m/z: calc'd for $C_{11}H_7BrClFN_2O_4S$: 364.9; found 365.9, 367.9 (M+1, M+3). ^{1}H NMR (300 MHz, MeOD) δ 7.09−7.20 (m, 2 H), 7.63 (d, J = 2.19 Hz, 1 H), 7.71 (d, J = 2.34 Hz, 1 H), 7.84−7.94 (m, 2 H).

*N-(2-Chloro-5-(4,4,5,5-tetramethyl-1,3,2-dioxaborolan-2-yl)-pyridin-3-yl)-4-fluorobenzenesulfonamide (**4b**).* This compound was prepared from **4a** (2.01 g, 5.51 mmol) using the general boronic ester formation method to afford the title compound (1.53 g, 67%) as a viscous oil, which solidified upon standing. MS (ESI pos. ion) m/z: calc'd for boronic acid $C_{11}H_9BClFN_2O_4S$: 330.0; found 331.0 (boronic acid M+1). ^{1}H NMR (300 MHz, $CDCl_3$) δ 1.33−1.41 (m, 12 H), 6.88 (s, 1 H), 7.10−7.19 (m, 2 H), 7.75−7.83 (m, 2 H), 8.31 (d, J = 1.6 Hz, 1 H), 8.46 (d, J = 1.6 Hz, 1 H).

*2-Chloro-1,5-naphthyridine (**6c**).* To a 100 mL round-bottomed flask was added 1,5-naphthyridine (**8**, 0.260 g, 2.00 mmol), DCM (10 mL), 3-chloroperoxybenzoic acid (0.517 g, 3.00 mmol). The reaction mixture was stirred at room temperature overnight. The solvent was removed in vacuo, and the residue was purified by silica gel chromatography (20% MeOH/EtOAc) to give 1,5-naphthyridine N-(1)-oxide (0.223 g, 76% yield). This material (0.198 g, 1.36 mmol) was dissolved in $POCl_3$ (2 mL) and the mixture was stirred at 100 °C for 8 h. The solvent was removed in vacuo. Saturated aqueous $NaHCO_3$ (2 mL) was added to the residue and the aqueous phase was extracted with EtOAc (2 × 20 mL). The combined organic phases were washed with saturated aqueous NaCl (2 mL), dried over sodium sulfate, filtered, and concentrated in vacuo. The crude product was purified by silica gel

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Table 5. Pharmacokinetic Properties of Selected Compounds

	in vitro microsomal stability[a]		in vivo rat PK[b]				
	rat	human	IV[c]			PO[d]	
Compd Number	CL μL/(min*mg)	CL μL/(min*mg)	CL (L/kg/h)	Vd,ss (L/kg)	MRT (h)	%F	AUC (ng*h/mL)
16d	108	47	0.17	0.44	3.0	92[e]	9554
16e	>399	247	–	–	–	–	–
16k	33	<14	0.13	0.81	6.3	73[f]	5630
16l	20	<14	0.04	0.52	15	70[e]	14700
18d	16	<14	0.02	0.19	9.0	88[e]	29560
18k	<14	25	0.59	1.21	2.1	16[f]	271
18q	32	31	0.08	0.40	5.4	72[e]	10750

[a] Single experimental value; estimated from parent compound (1 μM) remaining following a 30 min incubation in liver microsome (0.25 mg/mL) and NADPH (1 mM). [b] Pharmacokinetic parameters following administration in male Sprague−Dawley rat: 3 animals per study. [c] Dosed at 1 mg/kg as a solution in DMSO. [d] PO doses was 2 mg/kg for **16d** and were 1 mg/kg for all other compounds. [e] Dosed as a suspension in 1% Tween 80, 2% HPMC, pH 2.2 with methanesulfonic acid. [f] Dosed as a suspension in 1% Tween 80, 2% HPMC, pH 2.0 with HCl.

Table 6. Inhibition of AKT (Ser 473) Phosphorylation in a Mouse Liver Pharmacodynamic Model[a]

		0.3 mg/kg		1 mg/kg		3 mg/kg	
compd number	time (h)	% inh	conc[b] (ng/mL)	% inh	conc[b] (ng/mL)	% inh	conc[b] (ng/mL)
16d	3	65	145	75	492	97	1310
16k	3	70	210	97	899	99	2330
16l	3	89	326	98	950	>99	2470
18d	4	55	448	86	1730	97	4470
18q	4	50	155	86	417	95	1790

[a] % Inhibition was calculated using the level of AKT (Ser 473) phosphorylation of veh/HGF as 100%. [b] Total drug concentration in plasma.

chromatography (60% EtOAc/hexanes) to give the title compound (0.078 g, 35%). MS (ESI pos. ion) *m/z*: calc'd for $C_8H_5ClN_2$: 164.0; found 165.0 (M+1). ^{1}H NMR (300 MHz, $CDCl_3$) δ 7.64 (d, *J* = 8.77 Hz, 1 H), 7.66−7.72 (m, 1 H), 8.35 (t, *J* = 8.77 Hz, 2 H), 8.99 (dd, *J* = 4.17, 1.68 Hz, 1 H). Also isolated was 4-chloro-1,5-naphthyridine (**9**, 0.086 g, 39%). MS (ESI pos. ion) *m/z*: calc'd for $C_8H_5ClN_2$: 164.0; found 165.0. ^{1}H NMR (300 MHz, $CDCl_3$) δ 7.75 (dd, *J* = 8.55, 4.17 Hz, 1 H), 7.79 (d, *J* = 4.68 Hz, 1 H), 8.47 (dd, *J* = 8.62, 1.61 Hz, 1 H), 8.87 (d, *J* = 4.68 Hz, 1 H), 9.11 (dd, *J* = 4.09, 1.61 Hz, 1 H).

*1,7-Naphthyridin-6-yl trifluoromethanesulfonate (**6d**). (a). 3-(Cyanomethyl)picolinonitrile (**11**).* To a 100 mL round-bottomed flask was added 3-methylpicolinonitrile (**10**, 2.36 g, 20.0 mmol), NBS (7.82 g, 43.9 mmol), and CCl_4 (50 mL). The reaction mixture was stirred at reflux for 16 h and allowed to cool to room temperature. The resulting solid was removed and the filter cake was washed with 50% EtOAc/hexanes. The combined filtrate was concentrated in vacuo and the crude product was purified by silica gel chromatography (30% EtOAc/hexane) to give 3-(bromomethyl)picolinonitrile (2.56 g, 65% yield). To a solution of this 3-(bromomethyl)picolinonitrile (1.56 g, 7.92 mmol) in MeOH (50 mL), potassium cyanide (0.773 g, 1.19 mmol) was added. The reaction mixture was stirred at room temperature for 6 h. The solvent was removed in vacuo and the residue was dissolved in EtOAc (50 mL), washed with water (10 mL), saturated aqueous NaCl (10 mL), dried over sodium sulfate, filtered, and concentrated in vacuo. The crude product was purified by silica gel chromatography (40% EtOAc/hexanes) to give **11** (0.328 g, 29% yield). MS (ESI pos. ion) *m/z*: calc'd for $C_8H_5N_3$: 143.0; found 144.0 (M+1). ^{1}H NMR (300 MHz, $CDCl_3$) δ 4.06 (s, 2 H), 7.63 (dd, *J* = 8.11, 4.75 Hz, 1 H), 8.01−8.08 (m, 1 H), 8.73 (dd, *J* = 4.75, 1.39 Hz, 1 H).

*(b). 8-Bromo-1,7-naphthyridin-6-amine (**12**).* To a 50 mL round-bottomed flask was added hydrobromic acid, 30% in acetic acid (0.32 mL, 5.9 mmol), and compound **11** (0.280 g, 1.96 mmol) in AcOH (0.5 mL) at 0 °C. The reaction mixture was stirred at 0 °C for 30 min. The resulting solid was collected and washed with 50% EtOAc/hexanes, then saturated aqueous $NaHCO_3$ (5 mL) was added and the mixture was extracted with EtOAc (2 × 50 mL). The organic extract was washed with saturated aqueous NaCl (5 mL), dried over sodium sulfate, filtered, and concentrated in vacuo and the residue was purified by silica gel chromatography (40% EtOAc/hexanes) to give the title compound (0.312 g, 71%). MS (ESI pos. ion) *m/z*: calc'd for $C_8H_6BrN_3$: 223.0; found 224.0, 226.0 (M+1, M+3). ^{1}H NMR (300 MHz, $CDCl_3$) δ 4.63 (s, 2 H), 6.61 (s, 1 H), 7.42 (dd, *J* = 8.48, 4.09 Hz, 1 H), 7.85 (dd, *J* = 8.48, 1.61 Hz, 1 H), 8.78 (dd, *J* = 3.95, 1.61 Hz, 1 H).

*(c). 1,7-Naphthyridin-6-yl trifluoromethanesulfonate (**6d**).* To a 50 mL round-bottomed flask was added compound **12** (0.224 g, 1.00 mmol), potassium hydroxide (0.067 g, 1.2 mmol), 10% palladium on carbon (0.011 g, 0.1 mmol), and EtOH (2 mL). The mixture was stirred under a hydrogen balloon for 4 h. The mixture was filtered through a pad of Celite and washed with EtOAc. The filtrate was concentrated in vacuo and the crude product was purified by silica gel chromatography (100% EtOAc) to give 1,7-naphthyridin-6-amine (0.108 g, 74% yield). To a solution of this, 1,7-naphthyridin-6-amine (0.102 g, 0.70 mmol) in DMF (1.6 mL), sodium nitrite (0.097 g, 1.41 mmol), and trifluoromethanesulfonic acid (0.8 mL, 9.0 mmol) were added. The reaction mixture was stirred at room temperature for 2 h and then diluted with EtOAc (40 mL) and washed with water (5 mL), saturated $NaHCO_3$ (5 mL), and saturated NaCl (5 mL). The organic phase was dried over sodium sulfate, filtered, and concentrated in vacuo. The crude product was

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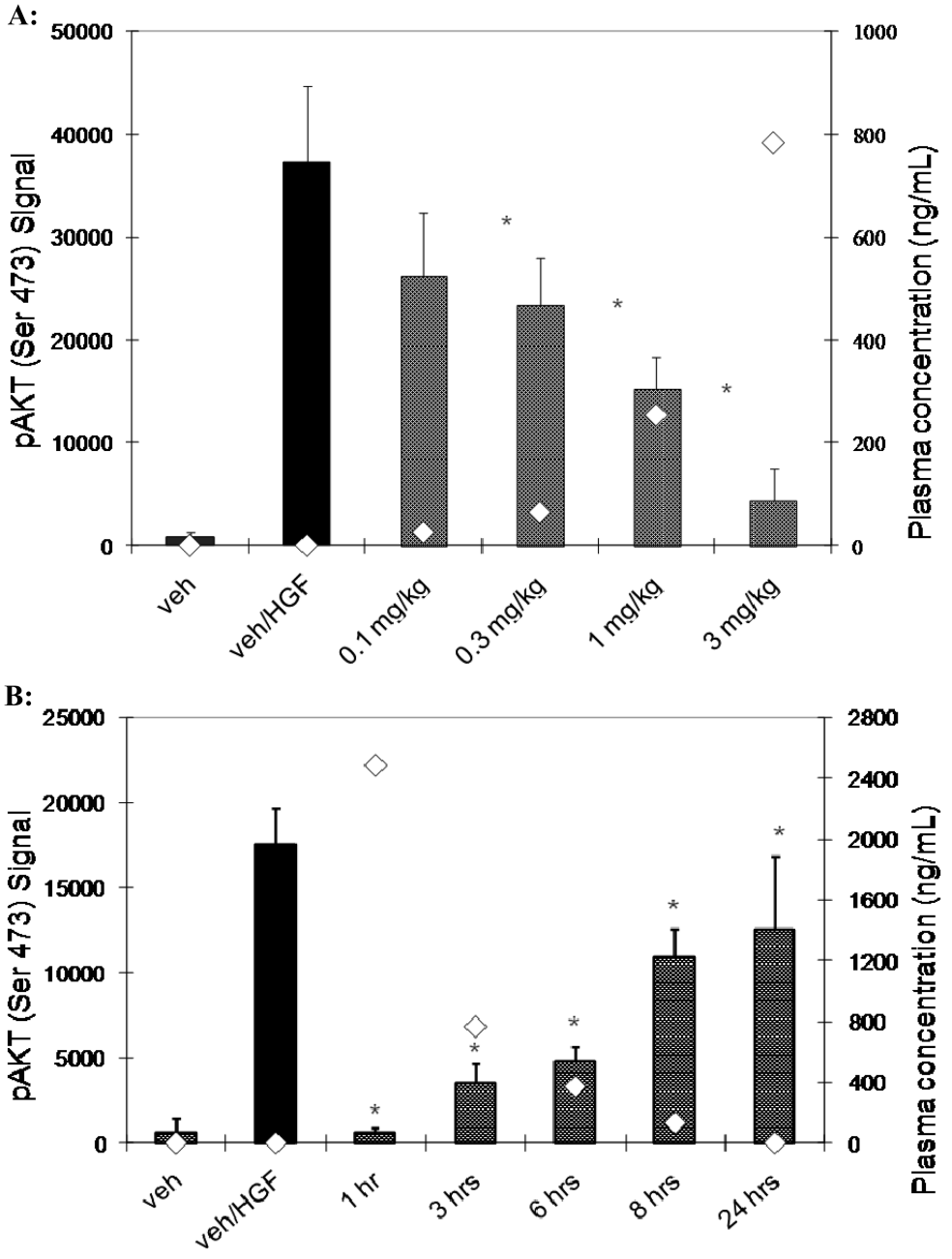

Figure 4. Effect of compound **16d** in a mouse liver pharmacodynamic model measuring the inhibition of HGF-stimulated AKT (Ser 473) phosphorylation:a (A) Dose−response study at 3 h post dose. (B) Time-course study at 3 mg/kg.

[a]Asterisks denote $p < 0.05$ compared with the vehicle/HGF group. Statistical significance was evaluated by Dunnett's method. Bars represent the average ± SD ($n = 3$). Diamonds represent mean plasma concentrations.

purified by silica gel chromatography (50% EtOAc/hexanes) to give the title compound (0.138 g, 71% yield) as a yellow solid. MS (ESI pos. ion) m/z: calc'd for $C_9H_5F_3N_2O_3S$: 278.0; found 279.0 (M+1). ^{1}H NMR (300 MHz, $CDCl_3$) δ 7.61 (s, 1 H), 7.71 (dd, J = 8.48, 4.24 Hz, 1 H), 8.21−8.30 (m, 1 H), 9.12 (dd, J = 4.24, 1.61 Hz, 1 H), 9.36 (s, 1 H).

*6-(4,4,5,5-Tetramethyl-1,3,2-dioxaborolan-2-yl)quinoxalines (**6f**).* This compound was prepared from 6-bromoquinoxaline (1.00 g, 4.78 mmol) using the general boronic ester formation method to afford the title compound (1.01 g, 82%) as a viscous amber oil. MS (ESI pos. ion) m/z: calc'd for $C_{14}H_{17}BN_2O_2$: 256.1; found 257.2 (M+1). ^{1}H NMR (400 MHz, $CDCl_3$) δ 1.40 (s, 12 H), 8.00−8.18 (m, 2 H), 8.60 (s, 1 H), 8.86 (d, J = 5.02 Hz, 2 H).

*N-(2-Chloro-5-(quinolin-6-yl)pyridin-3-yl)-4-fluorobenzenesulfonamide (**7a**).* This compound was prepared from **4a** (0.250 g, 0.684 mmol) and quinoline-6-boronic acid (0.177 g, 1.03 mmol) according to the general Suzuki coupling method to afford the title compound (0.140 g, 50%) as a white solid. MS (ESI pos. ion) m/z: calc'd for $C_{20}H_{13}ClFN_3O_2S$: 413.0; found 414.0 (M+1). HRMS calc'd for $C_{20}H_{13}ClFN_3O_2S$ (M + H) 414.0472: found; 414.0492. ^{1}H NMR (400 MHz, DMSO-d_6) δ 7.45 (t, J = 8.80 Hz, 2 H), 7.62 (dd, J = 8.22, 4.11 Hz, 1 H), 7.84 (dd,

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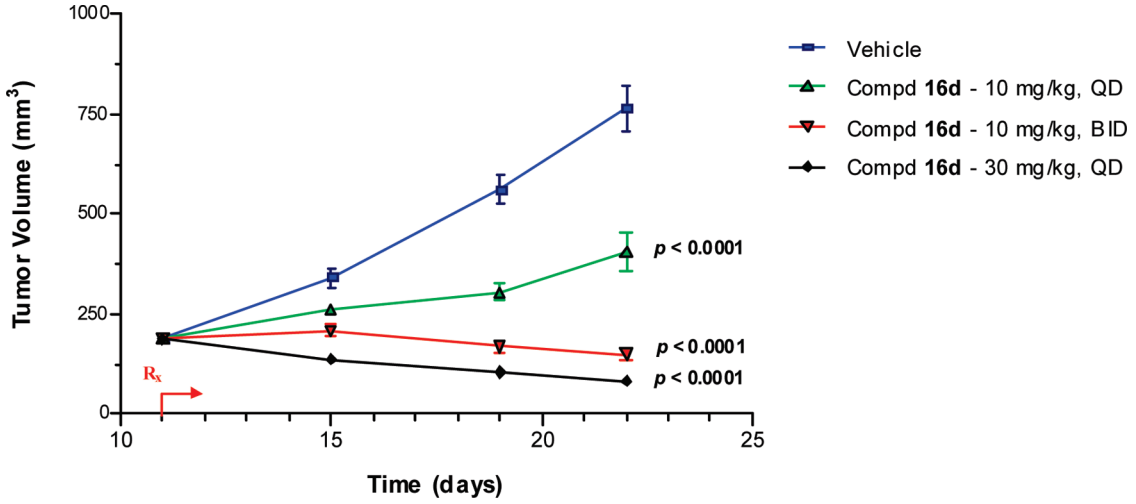

Figure 5. In vivo efficacy of compound **16d** in a PTEN Null U-87 MG glioblastoma xenograft model. Data represent the mean (n = 10) ± standard deviation. Statistical significance was evaluated by Repeated Measures ANOVA followed by Dunnett post hoc test.

J = 8.61, 5.09 Hz, 2 H), 8.05−8.11 (m, 1 H), 8.12−8.20 (m, 2 H), 8.35 (s, 1 H), 8.47 (d, J = 8.02 Hz, 1 H), 8.76 (d, J = 1.96 Hz, 1 H), 8.97 (d, J = 4.11 Hz, 1 H), 10.54 (s, 1 H).

*N-(2-Chloro-5-(isoquinolin-6-yl)pyridin-3-yl)-4-fluorobenzenesulfonamide (**7b**).* This compound was prepared from **4b** (0.262 g, 0.63 mmol) and 6-bromoisoquinoline (0.120 g, 0.58 mmol) according to the general Suzuki coupling method to afford the title compound (0.020 g, 8.4%) as a white solid. MS (ESI pos. ion) m/z: calc'd for $C_{20}H_{13}ClFN_3O_2S$: 413.0; found: 414.0 (M+1). HRMS calc'd for $C_{20}H_{13}ClFN_3O_2S$ (M + H): 414.0472; found 414.0483. ^{1}H NMR (400 MHz, DMSO-d_6) δ 7.44 (t, J = 8.80 Hz, 2 H), 7.82−7.85 (m, 2 H), 7.93 (d, J = 5.87 Hz, 1 H), 8.00 (dd, J = 8.61, 1.37 Hz, 1 H), 8.16 (d, J = 2.15 Hz, 1 H), 8.29 (d, J = 8.61 Hz, 1 H), 8.33 (s, 1 H), 8.58 (d, J = 5.67 Hz, 1 H), 8.75 (s, 1 H), 9.40 (s, 1 H), 10.58 (s, 1 H).

*N-(2-Chloro-5-(1,5-naphthyridin-2-yl)pyridin-3-yl)-4-fluorobenzenesulfonamide (**7c**).* This compound was prepared from **4b** (0.135 g, 0.33 mmol) and **6c** (0.054 g, 0.33 mmol) according to the general Suzuki coupling method to afford the title compound (0.078 g, 57%) as a white solid. MS (ESI pos. ion) m/z: calc'd for $C_{19}H_{12}ClFN_4O_2S$: 414.0; found 415.0 (M+1). HRMS calc'd for $C_{19}H_{12}ClFN_4O_2S$ (M + H): 415.0425; found 415.0420. ^{1}H NMR (300 MHz, $CDCl_3$) δ 7.07 (s, 1 H), 7.16 (t, J = 8.55 Hz, 2 H), 7.73 (dd, J = 8.55, 4.17 Hz, 1 H), 7.88 (dd, J = 8.92, 4.97 Hz, 2 H), 8.11 (d, J = 8.77 Hz, 1 H), 8.49 (d, J = 8.48 Hz, 1 H), 8.55 (d, J = 8.77 Hz, 1 H), 8.83 (d, J = 2.19 Hz, 1 H), 8.94 (d, J = 2.19 Hz, 1 H), 9.03 (dd, J = 4.09, 1.61 Hz, 1 H).

*N-(2-Chloro-5-(1,7-naphthyridin-6-yl)pyridin-3-yl)-4-fluorobenzenesulfonamide (**7d**).* This compound was prepared from **4b** (0.062 g, 0.15 mmol) and **6d** (0.042 g, 0.15 mmol) according to the general Suzuki coupling method to afford the title compound (0.046 g, 73%) as a white solid. MS (ESI pos. ion) m/z: calc'd for $C_{19}H_{12}ClFN_4O_2S$: 414.0; found 415.0 (M+1). HRMS calc'd for $C_{19}H_{12}ClFN_4O_2S$ (M + H): 415.0425; found 415.0430. ^{1}H NMR (300 MHz, $CDCl_3$) δ 7.01 (s, 1 H), 7.10−7.21 (m, 2 H), 7.68 (dd, J = 8.33, 4.24 Hz, 1 H), 7.82−7.91 (m, 2 H), 8.11 (d, J = 0.73 Hz, 1 H), 8.27 (d, J = 7.75 Hz, 1 H), 8.78 (d, J = 2.19 Hz, 1 H), 8.95 (d, J = 2.19 Hz, 1 H), 9.08 (dd, J = 4.17, 1.68 Hz, 1 H), 9.62 (s, 1 H).

*N-(2-Chloro-5-(1,8-naphthyridin-3-yl)pyridin-3-yl)-4-fluorobenzenesulfonamide (**7e**).* This compound was prepared from **4b** (0.140 g, 0.35 mmol) and 3-bromo-1,8-naphthyridine (0.073 g, 0.35 mmol) according to the general Suzuki coupling method to afford the title compound (0.098 g, 67%) as a white solid. MS (ESI pos. ion) m/z: calc'd for $C_{19}H_{12}ClFN_4O_2S$: 414.0; found 415.0 (M+1). HRMS calc'd for $C_{19}H_{12}ClFN_4O_2S$ (M + H): 415.0425; found 415.0410. ^{1}H NMR (300 MHz, MeOD) δ 7.30 (t, J = 8.77 Hz, 2 H), 7.76 (dd, J = 8.18, 4.38 Hz, 1 H), 7.89 (dd, J = 8.99, 5.04 Hz, 2 H), 8.44 (d, J = 2.34 Hz, 1 H), 8.62 (dd, J = 8.18, 1.90 Hz, 1 H), 8.70 (d, J = 2.34 Hz, 1 H), 8.79 (d, J = 2.48 Hz, 1 H), 9.15 (dd, J = 4.38, 1.90 Hz, 1 H), 9.39 (d, J = 2.48 Hz, 1 H).

*N-(2-Chloro-5-(quinoxalin-6-yl)pyridin-3-yl)-4-fluorobenzenesulfonamide (**7f**).* This compound was prepared from **4a** (0.300 g, 0.82 mmol) and **6f** (0.252 g, 0.98 mmol) according to the general Suzuki coupling method to afford the title compound (0.150 g, 44%) as a light-brown solid. MS (ESI pos. ion) m/z: calc'd for $C_{19}H_{12}ClFN_4O_2S$: 414.0; found 415.0 (M+1). HRMS calc'd for $C_{19}H_{12}ClFN_4O_2S$ (M + H): 415.0425; found 415.0410. ^{1}H NMR (400 MHz, DMSO-d_6) δ 7.44 (t, J = 8.78 Hz, 2 H), 7.84 (dd, J = 8.78, 5.27 Hz, 2 H), 8.13−8.29 (m, 3 H), 8.42 (s, 1 H), 8.83 (s, 1 H), 9.03 (d, J = 11.04 Hz, 2 H), 10.58 (s, 1 H).

*N-(2-Chloro-5-(quinazolin-6-yl)pyridin-3-yl)-4-fluorobenzenesulfonamide (**7g**).* This compound was prepared from **4b** (0.110 g, 0.27 mmol) and 6-bromoquinazoline (0.062 g, 0.30 mmol) according to the general Suzuki coupling method to afford the title compound (0.060 g, 54%) as a white solid. MS (ESI pos. ion) m/z: calc'd for $C_{19}H_{12}FClN_4O_2S$: 414.0; found 414.9 (M+1) observed in basic LC/MS condition (NH_4OH as additive). In acidic LC/MS condition with 0.1% TFA, the hydrate mass (M+19) was observed. HRMS calc'd for $C_{19}H_{12}ClFN_4O_2S$ (M + H): 415.0425; found 415.0410. ^{1}H NMR (400 MHz, DMSO-d_6) δ 7.45 (t, J = 8.80 Hz, 2 H), 7.84 (dd, J = 8.71, 5.18 Hz, 2 H), 8.17 (d, J = 8.80 Hz, 1 H), 8.23 (d, J = 2.35 Hz, 1 H), 8.38 (dd, J = 8.80, 1.76 Hz, 1 H), 8.55 (d, J = 1.37 Hz, 1 H), 8.78 (d, J = 2.15 Hz, 1 H), 9.37 (s, 1 H) 9.71 (s, 1 H), 10.59 (br s, 1 H).

*N-(2-Chloro-5-(cinnolin-6-yl)pyridin-3-yl)-4-fluorobenzenesulfonamide (**7h**).* This compound was prepared from **4b** (0.296 g, 0.72 mmol) and 6-bromocinnoline (0.150 g, 0.72 mmol) according to the general Suzuki coupling method to afford the title compound (0.015 g, 5%) as a yellow solid. MS (ESI pos. ion) m/z: calc'd for $C_{19}H_{12}ClFN_4O_2S$: 414.0; found 415.0 (M+1). HRMS calc'd for $C_{19}H_{12}ClFN_4O_2S$ (M + H): 415.0425; found 415.0410. ^{1}H NMR (400 MHz, DMSO-d_6) δ 7.44 (t, J = 8.78 Hz, 2 H), 7.83 (dd, J = 8.78, 5.27 Hz, 2 H), 8.24 (s, 1 H), 8.28−8.31 (m, 2 H), 8.46 (s, 1 H), 8.60 (d, J = 9.03 Hz, 1 H), 8.79 (s, 1 H), 9.46 (d, J = 5.52 Hz, 1 H), 10.60 (s, 1 H).

*4-Chloro-6-(4,4,5,5-tetramethyl-1,3,2-dioxaborolan-2-yl)quinoline (**15a**).* This compound was prepared from 6-bromo-4-chloroquinoline (1.5 g, 6.2 mmol) using the general boronic ester formation method to afford the title compound (1.45 g, 81%) as a brown solid. MS (ESI pos. ion) m/z: calc'd for $C_{15}H_{17}BClNO_2$: 289.1; found 290.3 (M+1). ^{1}H NMR

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(300 MHz, $CDCl_3$) δ 1.41 (s, 12 H), 7.51 (d, J = 4.82 Hz, 1 H), 8.05−8.12 (m, 1 H), 8.14 (s, 1 H), 8.73 (s, 1 H), 8.81 (d, J = 4.68 Hz, 1 H).

*6-Bromo-N,N-dimethylquinolin-4-amine (**15b**) and 6-bromo-N-(2-methoxyethyl)quinolin-4-amine (**15c**).* These compounds were prepared from 6-bromo-4-chloroquinoline (0.2 g, 0.8 mmol) and 2-methoxyethylamine (0.7 mL, 8 mmol) according to the general S_N2' reaction method to afford the following compounds. (i) 6-Bromo-*N,N*-dimethylquinolin-4-amine (**15b**) (0.150 g, 72%) as a yellow oil. MS (ESI pos. ion) m/z: calc'd for $C_{11}H_{11}BrN_2$: 250.0; found 251.0, 253.0 (M+1, M+3). ^{1}H NMR (300 MHz, $CDCl_3$) δ 3.03 (s, 6 H), 6.78 (d, J = 5.12 Hz, 1 H), 7.70 (dd, J = 8.92, 2.19 Hz, 1 H), 7.90 (d, J = 9.06 Hz, 1 H), 8.22 (d, J = 2.19 Hz, 1 H), 8.66 (d, J = 5.12 Hz, 1 H). (ii) 6-Bromo-*N*-(2-methoxyethyl)quinolin-4-amine (**15c**) (0.050 g, 22%) as a light-brown solid. MS (ESI pos. ion) m/z: calc'd for $C_{12}H_{13}BrN_2$: 280.0; found 281.0, 283.0 (M+1, M+3). ^{1}H NMR (300 MHz, $CDCl_3$) δ 3.42−3.55 (m, 5 H), 3.75 (t, J = 5.12 Hz, 2 H), 5.28 (br s, 1 H), 6.46 (d, J = 5.41 Hz, 1 H), 7.70 (dd, J = 9.06, 2.05 Hz, 1 H), 7.86 (d, J = 9.06 Hz, 1 H), 7.92 (d, J = 2.05 Hz, 1 H), 8.57 (d, J = 5.26 Hz, 1 H).

*6-Bromo-4-morpholinoquinoline (**15d**).* This compound was prepared from 6-bromo-4-chloroquinoline (3.0 g, 12 mmol) and morpholine (3.2 g, 37 mmol) according to the general S_N2' method to afford the title compound (3.4 g, 94%) as an off-white solid. MS (ESI pos. ion) m/z: calc'd for $C_{13}H_{13}BrN_2O$: 292.0; found: 292.9, 294.9 (M+1, M+3). ^{1}H NMR (300 MHz, $CDCl_3$) δ 3.17−3.26 (m, 4 H), 3.96−4.04 (m, 4 H), 6.89 (d, J = 4.97 Hz, 1 H), 7.73 (dd, J = 8.92, 2.19 Hz, 1 H), 7.94 (d, J = 8.92 Hz, 1 H), 8.16 (d, J = 2.19 Hz, 1 H), 8.76 (d, J = 4.97 Hz, 1 H).

*4-(4-(Pyridin-4-ylmethyl)piperazin-1-yl)quinolin-6-ylboronic Acid (**15e**).* This compound was prepared from **15a** (0.360 g, 1.24 mmol) and 1-(pyridin-4-ylmethyl)piperazine (0.500 g, 2.82 mmol) according to the general S_N2' method to afford the title compound (0.370 g, 86%) as a brown solid. MS (ES, pos.) m/z: calc'd for $C_{19}H_{21}BN_4O_2$: 348.2; found 349.2 (M+1).

*N-(2-Chloro-5-(4-chloroquinolin-6-yl)pyridin-3-yl)-4-fluorobenzenesulfonamide (**16a**).* This compound was prepared from **4a** (2 g, 5 mmol) and **15a** (2 g, 7 mmol) according to the general Suzuki coupling method to afford the title compound (1.5 g, 69%) as a light-yellow solid. MS (ESI pos. ion) m/z: calc'd for $C_{20}H_{12}Cl_2FN_3O_2S$: 447.5; found 448.3 (M+1). ^{1}H NMR (300 MHz, $CDCl_3$) δ 6.99 (br s, 1 H), 7.13−7.24 (m, 2 H), 7.60 (d, J = 4.68 Hz, 1 H), 7.88 (dd, J = 8.99, 4.90 Hz, 2 H), 7.97 (dd, J = 8.77, 2.05 Hz, 1 H), 8.28 (d, J = 8.77 Hz, 1 H), 8.33 (d, J = 2.19 Hz, 1 H), 8.38 (d, J = 1.90 Hz, 1 H), 8.52 (d, J = 2.34 Hz, 1 H), 8.87 (d, J = 4.68 Hz, 1 H).

*N-(2-Chloro-5-(4-(dimethylamino)quinolin-6-yl)pyridin-3-yl)-4-fluorobenzenesulfonamide (**16b**).* This compound was prepared from **4b** (0.1 g, 0.3 mmol) and **15b** (0.07 g, 0.3 mmol) according to the general Suzuki coupling method to afford the title compound (0.055 g, 43%) as a light-yellow solid. MS (ESI pos. ion) m/z: calc'd for $C_{22}H_{18}ClFN_4O_2S$: 456.1; found 457.0 (M+1). HRMS calc'd for $C_{22}H_{18}ClFN_4O_2S$ (M + H): 457.0893; found 457.0890. ^{1}H NMR (300 MHz, $CDCl_3$) δ 3.12 (s, 6 H), 6.85 (d, J = 5.12 Hz, 1 H), 7.18 (t, J = 8.55 Hz, 2 H), 7.84 (td, J = 8.66, 3.58 Hz, 3 H), 8.16 (d, J = 8.77 Hz, 1 H), 8.24 (d, J = 2.05 Hz, 1 H), 8.32 (d, J = 2.19 Hz, 1 H), 8.50 (d, J = 2.34 Hz, 1 H), 8.72 (d, J = 5.12 Hz, 1 H).

*N-(2-Chloro-5-(4-(2-methoxyethylamino)quinolin-6-yl)pyridin-3-yl)-4-fluorobenzenesulfonamide (**16c**).* This compound was prepared from **4b** (0.07 g, 0.2 mmol) and **15c** (0.05 g, 0.2 mmol) according to the general Suzuki coupling method to afford the title compound (0.015 g, 17%) as a light-yellow solid. MS (ESI pos. ion) m/z: calc'd for $C_{23}H_{20}ClFN_4O_3S$: 486.1; found: 487.0 (M+1). HRMS calc'd for $C_{23}H_{20}ClFN_4O_3S$ (M + H): 487.0998; found 487.1000. ^{1}H NMR (300 MHz, MeOD) δ 3.41 (s, 3 H), 3.76 (s, 4 H), 6.87 (d, J = 6.87 Hz, 1 H), 7.13−7.25 (m, 2 H), 7.83−7.92 (m, 2 H), 7.94 (s, 1 H), 8.02−8.10 (m, 1 H), 8.20 (s, 1 H), 8.32−8.42 (m, 2 H), 8.59 (s, 1 H).

*N-(2-Chloro-5-(4-morpholinoquinolin-6-yl)pyridin-3-yl)-4-fluorobenzenesulfonamide (**16d**).* This compound was prepared from **4b** (2.01 g, 4.87 mmol) and **15d** (1.58 g, 5.38 mmol) according to the general Suzuki coupling method to afford the title compound (1.71 g, 70%) as an off-white solid. MS (ESI pos. ion) m/z: calc'd for $C_{24}H_{20}ClFN_4O_3S$: 498.1; found 499.0 (M+1). HRMS calc'd for $C_{24}H_{20}ClFN_4O_3S$ (M + H): 499.0998; found 499.1010. ^{1}H NMR (400 MHz, DMSO-d_6) δ 3.27−3.33 (m, 4 H), 3.87−3.93 (m, 4 H), 7.09 (d, J = 5.28 Hz, 1 H), 7.42 (t, J = 8.80 Hz, 2 H), 7.79−7.87 (m, 2 H), 7.99−8.04 (m, 1 H), 8.06−8.12 (m, 2 H), 8.21 (d, J = 1.76 Hz, 1 H), 8.65 (d, J = 2.15 Hz, 1 H), 8.75 (d, J = 5.28 Hz, 1 H), 10.87 (br s, 1 H).

*N-(2-Chloro-5-(4-(4-(pyridin-4-ylmethyl)piperazin-1-yl)quinolin-6-yl)pyridin-3-yl)-4-fluorobenzenesulfonamide (**16e**).* This compound was prepared from **4a** (0.400 g, 1.1 mmol) and **15e** (0.370 g, 1.06 mmol) according to the general Suzuki coupling method to afford the title compound (0.320 g, 51% yield) as a yellow solid. MS (ES, pos.) m/z: calc'd for $C_{30}H_{26}ClFN_6O_2S$ 588.2; found 589.1(M+1). HRMS calc'd for $C_{30}H_{26}ClFN_6O_2S$ (M + H): 589.1579; found 589.1582. ^{1}H NMR (400 MHz, DMSO-d_6) δ 2.74 (br s, 4 H), 3.35 (br s, 4 H, overlapped with water), 3.68 (s, 2 H), 7.08 (d, J = 5.09 Hz, 1 H), 7.35−7.45 (m, 4 H), 7.83 (dd, J = 8.71, 5.18 Hz, 2 H), 7.97−8.03 (m, 1 H), 8.05−8.13 (m, 3 H), 8.17 (s, 1 H), 8.54 (d, J = 4.70 Hz, 2 H), 8.61 (d, J = 1.76 Hz, 1 H), 8.73 (d, J = 5.09 Hz, 1 H), 10.94 (br s, 1H).

*N-(2-Chloro-5-(4-(piperidin-1-yl)quinolin-6-yl)pyridin-3-yl)-4-fluorobenzenesulfonamide (**16f**).* This compound was prepared from **15a** (0.07 g, 0.2 mmol) and piperidine (0.05 mL, 0.5 mmol) according to the general S_N2' method to afford the title compound (0.04 g, 52%) as a white solid. MS (ESI pos. ion) m/z: calc'd for $C_{25}H_{22}ClFN_4O_2S$ 496.1; found 497.5 (M+1). HRMS calc'd for $C_{25}H_{22}ClFN_4O_2S$ (M + H): 497.1205; found 497.1210. ^{1}H NMR (300 MHz, $CDCl_3$) δ 1.67−1.79 (m, 2 H) 1.83−1.96 (m, 4 H) 3.17−3.31 (m, 4 H) 6.88 (d, J = 4.97 Hz, 1 H) 7.11−7.21 (m, 2 H) 7.77−7.90 (m, 3 H) 8.16 (s, 1 H) 8.21 (d, J = 2.05 Hz, 1 H) 8.34 (d, J = 2.34 Hz, 1 H) 8.49 (s, 1 H) 8.74 (d, J = 5.12 Hz, 1 H).

*N-(2-Chloro-5-(4-(4-hydroxypiperidin-1-yl)quinolin-6-yl)pyridin-3-yl)-4-fluorobenzenesulfonamide (**16g**).* This compound was prepared from **15a** (0.08 g, 0.2 mmol) and 4-hydroxypiperidine (0.2 g, 2 mmol) according to the general S_N2' method to afford the title compound (0.035 g, 38%) as a white solid. MS (ESI pos. ion) m/z: calc'd for $C_{25}H_{22}ClFN_4O_3S$: 493.1; found 494.3 (M+1). HRMS calc'd for $C_{25}H_{22}ClFN_4O_3S$ (M + H): 513.1154; found 513.1130. ^{1}H NMR (300 MHz, $CDCl_3$) δ 1.83−2.02 (m, 2 H), 2.21 (ddd, J = 6.58, 3.22, 3.07 Hz, 2 H), 3.20−3.38 (m, 2 H), 3.56−3.78 (m, 2 H), 4.12 (ddd, J = 8.26, 5.85, 2.85 Hz, 1 H), 6.96 (d, J = 5.55 Hz, 1 H), 7.19 (t, J = 8.55 Hz, 2 H), 7.85 (dd, J = 9.06, 4.97 Hz, 3 H), 7.88−7.94 (m, 1 H), 8.19 (s, 1 H), 8.34 (d, J = 2.19 Hz, 1 H), 8.49 (d, J = 2.34 Hz, 1 H), 8.71 (d, J = 5.70 Hz, 1 H).

*N-(2-Chloro-5-(4-(4-isopropylpiperazin-1-yl)quinolin-6-yl)pyridin-3-yl)-4-fluorobenzenesulfonamide (**16h**).* This compound was prepared from **15a** (0.07 g, 0.2 mmol) and 1-isopropylpiperazine (0.1 mL, 0.8 mmol) according to the general S_N2' method to afford the title compound (0.055 g, 65%) as a white solid. MS (ESI pos. ion) m/z: calc'd for $C_{27}H_{27}ClFN_5O_2S$ 539.2; found 540.3 (M+1). HRMS calc'd for $C_{27}H_{27}ClFN_5O_2S$ (M + H): 540.1626; found 540.1650. ^{1}H NMR (300 MHz, $CDCl_3$) δ 1.15 (d, J = 6.58 Hz, 6 H), 2.75−2.95 (m, 5 H), 3.27−3.39 (m, 4 H), 6.93 (d, J = 4.97 Hz, 1 H), 7.18 (t, J = 8.55 Hz, 2 H), 7.77−7.92 (m, 3 H), 8.19 (d, J = 8.62 Hz, 1 H), 8.27 (d, J = 1.90 Hz, 1 H), 8.37 (d, J = 2.34 Hz, 1 H), 8.51 (d, J = 2.34 Hz, 1 H), 8.79 (d, J = 4.97 Hz, 1 H).

*N-(5-(4-(4-Benzylpiperazin-1-yl)quinolin-6-yl)-2-chloropyridin-3-yl)-4-fluorobenzenesulfonamide (**16i**).* This compound was prepared from **15a** (0.05 g, 0.1 mmol) and 1-benzylpiperazine (0.2 g, 1 mmol) according to the general S_N2' method to afford the title compound (0.035 g, 53%) as a white solid. MS (ESI pos. ion) m/z: calc'd for $C_{31}H_{27}ClFN_5O_2S$ 587.2; found 588.3 (M+1). HRMS calc'd for $C_{31}H_{27}ClFN_5O_2S$ (M + H): 588.1626; found 588.1630. ^{1}H NMR (300 MHz, $CDCl_3$) δ 2.81 (br s, 4 H), 3.33 (br s, 4 H), 3.67 (s, 2 H), 6.92 (d, J = 5.12 Hz, 1 H), 7.17 (t, J = 8.48 Hz, 2 H), 7.29−7.44 (m, 5 H), 7.85

(dd, J = 8.92, 4.82 Hz, 3 H), 8.18 (d, J = 8.77 Hz, 1 H), 8.25 (s, 1 H), 8.35 (d, J = 2.34 Hz, 1 H), 8.50 (d, J = 2.34 Hz, 1 H), 8.78 (d, J = 4.97 Hz, 1 H).

*N-(2-Chloro-5-(4-phenylquinolin-6-yl)pyridin-3-yl)-4-fluorobenzenesulfonamide (**16j**).* This compound was prepared from **15a** (0.07 g, 0.2 mmol) and phenylboronic acid (0.06 g, 0.5 mmol) according to the general Suzuki coupling method to afford the title compound (0.030 g, 39%) as a light-yellow solid. MS (ESI pos. ion) m/z: calc'd for $C_{26}H_{17}ClFN_3O_2S$: 489.1; found 490.0 (M+1). HRMS calc'd for $C_{26}H_{17}ClFN_3O_2S$ (M + H): 490.0784; found 490.0770. ^{1}H NMR (300 MHz, $CDCl_3$) δ 7.01−7.12 (m, 2 H), 7.44 (d, J = 4.53 Hz, 1 H), 7.54−7.64 (m, 5 H), 7.78 (dd, J = 9.06, 4.97 Hz, 2 H), 7.92 (dd, J = 8.77, 2.19 Hz, 1 H), 8.08 (d, J = 1.75 Hz, 1 H), 8.20 (d, J = 2.34 Hz, 1 H), 8.32 (d, J = 8.62 Hz, 1 H), 8.37 (d, J = 2.19 Hz, 1 H), 9.02 (d, J = 4.53 Hz, 1 H).

*N-(2-Chloro-5-(4-(pyridin-3-yl)quinolin-6-yl)pyridin-3-yl)-4-fluorobenzenesulfonamide (**16k**).* To a 5 mL vial was added **15a** (0.100 g, 0.2 mmol), 3-(4,4,5,5-tetramethyl-1,3,2-dioxaborolan-2-yl)pyridine (0.05 g, 0.3 mmol), potassium acetate (0.05 g, 0.6 mmol), dichlorobis(di-*tert*-butylphenylphosphine)palladium(II) (0.007 g, 0.01 mmol), water (0.3 mL), and 1-butanol (3 mL). The reaction vial was heated at 100 °C for 2 h. The solvent was removed and the residue was partitioned between water and $CHCl_3$. The organic layer was dried over magnesium sulfate and concentrated in vacuo. The crude product was purified by silica gel chromatography (DCM/EtOAc/MeOH = 75:22:3) to afford the title compound (0.070 g, 64%) as a white solid. MS (ESI pos. ion) m/z: calc'd for $C_{25}H_{16}ClFN_4O_2S$ 490.1; found 490.9 (M+1). HRMS calc'd for $C_{25}H_{16}ClFN_4O_2S$ (M + H): 491.0734; found 491.0730. ^{1}H NMR (300 MHz, $CDCl_3$) δ 7.01−7.07 (m, 1 H), 7.11−7.21 (m, 2 H), 7.47 (d, J = 4.53 Hz, 1 H), 7.58 (d, J = 3.07 Hz, 1 H), 7.74−7.84 (m, 2 H), 7.89−8.00 (m, 3 H), 8.20 (d, J = 2.34 Hz, 1 H), 8.34−8.42 (m, 2 H), 8.79−8.88 (m, 2 H), 9.07 (d, J = 4.38 Hz, 1 H).

*N-(2-Chloro-5-(4-(pyridin-4-yl)quinolin-6-yl)pyridin-3-yl)-4-fluorobenzenesulfonamide (**16l**).* This compound was prepared from **15a** (0.1 g, 0.2 mmol) and 4-(4,4,5,5-tetramethyl-1,3,2-dioxaborolan-2-yl)pyridine (0.05 g, 0.3 mmol) in a similar manner as described for **16k** to afford the title compound (0.07 g, 64%) as a white solid. MS (ESI pos. ion) m/z: calc'd for $C_{25}H_{16}ClFN_4O_2S$ 490.1; found 490.9 (M+1). HRMS calc'd for $C_{25}H_{16}ClFN_4O_2S$ (M + H): 491.0734; found 491.0740. ^{1}H NMR (300 MHz, $CDCl_3$) δ 7.04−7.19 (m, 3 H), 7.44 (d, J = 4.38 Hz, 1 H), 7.51 (dd, J = 4.46, 1.53 Hz, 2 H), 7.72−7.84 (m, 2 H) 7.92−8.02 (m, 2 H), 8.22 (d, J = 2.34 Hz, 1 H), 8.31−8.40 (m, 2 H), 8.87 (dd, J = 4.38, 1.61 Hz, 2 H), 9.07 (d, J = 4.38 Hz, 1 H).

*N-(2-Chloro-5-(4-(4-(dimethylamino)phenyl)quinolin-6-yl)pyridin-3-yl)-4-fluorobenzenesulfonamide (**16m**).* This compound was prepared from **15a** (0.07 g, 0.2 mmol) and *N*,*N*-dimethyl-4-(4,4,5,5-tetramethyl-1,3,2-dioxaborolan-2-yl)benzenamine (0.05 g, 0.2 mmol) in a similar manner to prepare **16k** to afford the title compound (0.035 g, 42%) as a red solid. MS (ESI pos. ion) m/z: calc'd for $C_{28}H_{22}ClFN_4O_2S$ 532.1; found 532.9 (M+1). HRMS calc'd for $C_{28}H_{22}ClFN_4O_2S$ (M + H): 533.1205; found 533.1190. ^{1}H NMR (300 MHz, $CDCl_3$) δ 3.07 (s, 6 H), 6.88−6.97 (m, 2 H), 7.04−7.13 (m, 3 H), 7.42 (d, J = 4.53 Hz, 1 H), 7.47−7.54 (m, 2 H), 7.77−7.84 (m, 2 H), 7.90 (dd, J = 8.77, 2.05 Hz, 1 H), 8.22−8.26 (m, 2 H), 8.28 (d, J = 8.77 Hz, 1 H), 8.41 (d, J = 2.34 Hz, 1 H), 8.96 (d, J = 4.53 Hz, 1 H).

*7-Bromo-2-chloroquinoxaline (**17a**).* To a stirred solution of quinoxalin-2-ol (10.0 g, 68.4 mmol) in 500 mL of AcOH in 1-L round-bottomed flask, bromine (3.60 mL, 70.3 mmol) was added slowly. The mixture was stirred at room temperature for 1.5 h. The resulting precipitate was collected via filtration and was washed with water and MeOH. This solid was dissolved into DMSO and water was added to create a precipitate, which was collected via filtration and washed with water and MeOH to afford 7-bromoquinoxalin-2-ol (3.85 g) as a light-pink solid. To the first filtrate was added water (1 L) and the resulting precipitate was collected and washed with copious amounts of water to afford another batch of 7-bromoquinoxalin-2-ol (7.26 g) as an off-white solid. The combined yield of this reaction was 11.11 g, 72%. To this 7-bromoquinoxalin-2-ol (8.01 g, 35.6 mmol) was added toluene (70 mL) and $POCl_3$ (14.9 mL, 160 mmol) and the mixture was stirred at room temperature for overnight. The reaction mixture was poured into ice−water (200 mL). The aqueous phase was extracted with EtOAc (3 × 150 mL). The combined organic phases were washed with saturated aqueous NaCl (400 mL), dried over sodium sulfate, filtered, and concentrated. Upon addition of DCM to the residue, some of the precipitated product was collected via filtration to afford the title compound as a light-brown powder (3.07 g). The filtrate containing the rest of the product was purified by silica gel column chromatography (0−8% EtOAc in hexanes) to afford more of the title compound (5.20 g) as a pink solid. Total yield was 8.27 g, 95%. MS (ESI pos. ion) m/z: calc'd for $C_8H_4BrClN_2$: 241.9; found 242.9, 244.9 (M+1, M+3). ^{1}H NMR (300 MHz, $CDCl_3$) δ 7.83−7.90 (m, 1 H), 7.95−8.02 (m, 1 H), 8.21 (d, J = 2.05 Hz, 1 H), 8.78 (s, 1 H).

*7-Bromo-2-fluoroquinoxaline (**17b**).* To a 150 mL round-bottomed flask, **7a** (1.73 g, 7.12 mmol) was suspended in DMSO (20 mL). Tetrabutylammonium fluoride (1.0 M in THF, 8.55 mL, 8.55 mmol) was added. The reaction mixture was stirred at room temperature for 1.5 h. Water (100 mL) was added and the resulting solid was collected, washed with water, and air-dried to obtain the title compound (1.54 g, 95%) as a light-yellow solid. This compound was not detectable in mass spectroscopy. ^{1}H NMR (300 MHz, DMSO-d_6) δ 7.92−8.41 (m, 3 H), 8.90−9.14 (m, 1 H).

*7-Bromo-N,N-dimethylquinoxalin-2-amine (**17c**).* This compound was prepared from **17b** (0.113 g, 0.499 mmol) and dimethylamine (2 M in THF, 1.0 mL, 2.00 mmol) according to the general S_N2' method to afford the title compound (0.124 g, 99%) as a pale-yellow solid. MS (ESI, pos. ion) m/z: calc'd for $C_{10}H_{10}BrN_3$: 251.0; found 251.9, 253.9 (M+1, M+3). ^{1}H NMR (400 MHz, $CDCl_3$) δ 3.27 (s, 6 H), 7.42 (dd, J = 8.71, 2.05 Hz, 1 H), 7.71 (d, J = 8.61 Hz, 1 H), 7.86 (d, J = 2.15 Hz, 1 H), 8.49 (s, 1 H).

*7-Bromo-2-morpholinoquinoxaline (**17d**).* This compound was prepared from **17c** (0.201 g, 0.826 mmol) and morpholine (0.200 mL, 2.29 mmol) according to the general S_N2' method to afford the title compound (0.124 g, 51%) as light-yellow crystals. MS (ESI, pos. ion) m/z: calc'd for $C_{12}H_{12}BrN_3O$ 293.0; found 294.0, 295.9 (M+1, M+3). ^{1}H NMR (300 MHz, $CDCl_3$) δ 3.72−3.83 (m, 4 H), 3.83−3.93 (m, 4 H), 7.49 (dd, J = 8.8, 2.2 Hz, 1 H), 7.75 (d, J = 8.6 Hz, 1 H), 7.88 (d, J = 2.0 Hz, 1 H), 8.55 (s, 1 H).

*N-(2-Chloro-5-(3-chloroquinoxalin-6-yl)pyridin-3-yl)-4-fluorobenzenesulfonamide (**18a**).* This compound was prepared from **4b** (0.189 g, 0.457 mmol) and **17a** (0.100 g, 0.410 mmol) according to the general Suzuki coupling method, except bis(di-*tert*-butylphosphine)-dichloropalladium(II) was used as a catalyst, to afford the title compound (0.126 g, 68%) as an off-white solid. MS (ESI pos. ion) m/z: calc'd for $C_{19}H_{11}Cl_2FN_4O_2S$ 448.0; found 448.8, 450.8 (M+1, M+3). ^{1}H NMR (300 MHz, DMSO-d_6) δ 7.38−7.50 (m, 2 H), 7.79−7.89 (m, 2 H), 8.18−8.35 (m, 3 H), 8.40 (d, J = 1.75 Hz, 1 H), 8.80 (d, J = 2.34 Hz, 1 H), 9.05 (s, 1 H), 10.56 (br s, 1 H).

*N-(2-Chloro-5-(3-(dimethylamino)quinoxalin-6-yl)pyridin-3-yl)-4-fluorobenzenesulfonamide (**18c**).* This compound was prepared from **4b** (0.161 g, 0.389 mmol) and **17c** (0.0952 g, 0.378 mmol) according to the general Suzuki coupling method to afford the title compound (0.136 g, 79%) as a yellow solid. MS (ESI, pos. ion) m/z: calc'd for $C_{21}H_{17}ClFN_5O_2S$ 457.1; found 457.9 (M+1). HRMS calc'd for $C_{21}H_{17}ClFN_5O_2S$ (M + H): 458.0846; found 458.0870. ^{1}H NMR (400 MHz, DMSO-d_6) δ 3.27 (s, 6 H), 7.45 (t, J = 8.80 Hz, 2 H), 7.64 (dd, J = 8.61, 1.76 Hz, 1 H), 7.78−7.88 (m, 3 H), 7.93 (d, J = 8.41 Hz, 1 H), 8.05 (d, J = 2.15 Hz, 1 H), 8.74 (s, 2 H), 10.52 (br s, 1 H).

*N-(2-Chloro-5-(3-morpholinoquinoxalin-6-yl)pyridin-3-yl)-4-fluorobenzenesulfonamide (**18d**).* This compound was prepared from **4b** (2.00 g, 4.85 mmol) and **17d** (1.58 g, 5.36 mmol) according to the general Suzuki coupling method to afford the title compound (1.07 g,

44%) as a pale-yellow solid. MS (ESI, pos. ion) m/z: calc'd for $C_{23}H_{19}ClFN_5O_3S$ 499.1; found 500.0 (M+1). HRMS calc'd for $C_{23}H_{19}ClFN_5O_2S$ (M + H): 500.0951; found 500.0960. ^{1}H NMR (300 MHz, DMSO-d_6) δ 3.67−3.91 (m, 8 H), 7.37−7.52 (m, 2 H), 7.70 (dd, J = 8.6, 2.0 Hz, 1 H), 7.76−7.89 (m, 3 H), 7.96 (d, J = 8.5 Hz, 1 H), 8.06 (d, J = 2.3 Hz, 1 H), 8.72 (d, J = 2.2 Hz, 1 H), 8.87 (s, 1 H), 10.53 (s, 1 H).

*N-(2-Chloro-5-(3-(pyridin-4-yl)quinoxalin-6-yl)pyridin-3-yl)-4-fluorobenzenesulfonamide (**18e**).* This compound was prepared from **18a** (0.106 g, 0.236 mmol) and pyridine-4-ylboronic acid (0.043 g, 0.35 mmol) according to the general Suzuki coupling method, except bis(di-*t*-butylphosphine)dichloropalladium(II) was used as the catalyst and potassium acetate was used for the base, to afford the title compound (0.066 g, 57%) as a light-yellow solid. MS (ESI pos. ion) m/z: calc'd for $C_{24}H_{15}ClFN_5O_2S$ 491.1; found 491.9 (M+1). HRMS calc'd for $C_{24}H_{15}ClFN_5O_2S$ (M + H): 492.0690; found 492.0700. ^{1}H NMR (300 MHz, $CDCl_3$) δ 7.06−7.23 (m, 3 H), 7.82−7.92 (m, 2 H), 8.01 (dd, J = 8.77, 2.05 Hz, 1 H), 8.14 (dd, J = 4.53, 1.61 Hz, 2 H), 8.31 (d, J = 8.77 Hz, 1 H), 8.40 (t, J = 1.83 Hz, 2 H), 8.57 (d, J = 2.34 Hz, 1 H), 8.89 (dd, J = 4.68, 1.46 Hz, 2 H), 9.42 (s, 1 H).

*General Method for Library Synthesis of **18f**−**18k**, **18m**−**18p**, and **18r**.* To a solution of **18b** (40 mg, 92 μmol) in DMSO (1 mL) was added the amine (1 M in DMF, 231 μL, 231 μmol) and DMSO (0.57 mL). The reaction was heated at 60 °C for 24 h. The reaction was then cooled to 23 °C and submitted to mass-directed purification to give the product as a yellow solid. The yield and spectroscopic data are shown below.

*N-(2-Chloro-5-(3-(2-methoxyethylamino)quinoxalin-6-yl)pyridin-3-yl)-4-fluorobenzenesulfonamide (**18f**).* 6.4 mg. Yield 11%. MS (ESI pos. ion) m/z: calc'd for $C_{22}H_{19}ClFN_5O_3S$ 487.1; found 488.0 (M+1). HRMS calc'd for $C_{22}H_{19}ClFN_5O_3S$ (M + H) 488.0951, found 488.0950. ^{1}H NMR (300 MHz, $CDCl_3$) δ 3.44 (s, 3 H), 3.70 (d, J = 4.82 Hz, 2 H), 3.80 (d, J = 5.55 Hz, 2 H), 7.16 (t, J = 8.62 Hz, 2 H), 7.51−7.62 (m, 2 H), 7.81−7.89 (m, 3 H), 7.98 (d, J = 8.62 Hz, 1 H), 8.25 (s, 1 H), 8.33 (d, J = 2.19 Hz, 1 H), 8.50 (d, J = 2.19 Hz, 1 H).

*N-(2-Chloro-5-(3-((2-methoxyethyl)(methyl)amino)quinoxalin-6-yl)-pyridin-3-yl)-4-fluorobenzenesulfonamide (**18g**).* 14.1 mg. Yield 31%. MS (ESI pos. ion) m/z: calc'd for $C_{23}H_{21}ClFN_5O_3S$ 501.1; found 502.0 (M+1). HRMS calc'd for $C_{23}H_{21}ClFN_5O_3S$ (M + H): 502.1107; found 502.1120. ^{1}H NMR (300 MHz, $CDCl_3$) δ 3.36 (s, 3 H), 3.39 (s, 3 H), 3.69−3.75 (m, 2 H), 3.94 (t, J = 5.33 Hz, 2 H), 7.01 (s, 1 H), 7.18 (t, J = 8.55 Hz, 2 H), 7.53 (dd, J = 8.48, 2.05 Hz, 1 H), 7.81−7.89 (m, 3 H), 7.99 (d, J = 8.48 Hz, 1 H), 8.33 (d, J = 2.19 Hz, 1 H), 8.51 (d, J = 2.34 Hz, 1 H), 8.61 (s, 1 H).

*N-(2-Chloro-5-(3-(cyclohexylmethylamino)quinoxalin-6-yl)pyridin-3-yl)-4-fluorobenzenesulfonamide (**18h**).* 16.9 mg. Yield 29%. MS (ESI pos. ion) m/z: calc'd for $C_{26}H_{25}ClFN_5O_2S$ 525.1; found 526.0 (M+1). HRMS calc'd for $C_{26}H_{25}ClFN_5O_2S$ (M + H): 526.1470; found 526.1480. ^{1}H NMR (300 MHz, $CDCl_3$) δ 1.00−1.38 (m, 5 H), 1.66−1.93 (m, 6 H), 3.45 (t, J = 6.21 Hz, 2 H), 7.05 (br s, 1 H), 7.18 (t, J = 8.55 Hz, 2 H), 7.55 (dd, J = 8.40, 1.97 Hz, 1 H), 7.82−7.90 (m, 3 H), 7.98 (d, J = 8.48 Hz, 1 H), 8.31 (d, J = 2.19 Hz, 2 H), 8.48 (d, J = 2.19 Hz, 1 H).

*N-(5-(3-(Benzyl(methyl)amino)quinoxalin-6-yl)-2-chloropyridin-3-yl)-4-fluorobenzenesulfonamide (**18i**).* 9.3 mg. Yield 31%. MS (ESI pos. ion) m/z: calc'd for $C_{27}H_{21}ClFN_5O_2S$ 533.1; found 534.0 (M+1). ^{1}H NMR (300 MHz, $CDCl_3$) δ 3.33 (s, 3 H), 4.99 (s, 2 H), 7.01 (s, 1 H), 7.11−7.25 (m, 2 H), 7.27−7.38 (m, 5 H), 7.54 (dd, J = 8.48, 2.05 Hz, 1 H), 7.81−7.92 (m, 3 H), 7.99 (d, J = 8.48 Hz, 1 H), 8.32 (d, J = 2.19 Hz, 1 H), 8.48−8.57 (m, 2 H).

*N-(2-Chloro-5-(3-(phenethylamino)quinoxalin-6-yl)pyridin-3-yl)-4-fluorobenzenesulfonamide (**18j**).* 20.8 mg. Yield 35%. MS (ESI pos. ion) m/z: calc'd for $C_{27}H_{21}ClFN_5O_2S$ 533.1; found 534.0(M+1). ^{1}H NMR (300 MHz, $CDCl_3$) δ 3.11 (t, J = 6.87 Hz, 2 H), 3.90 (d, J = 4.97 Hz, 2 H), 7.11 (br s, 1 H), 7.18−7.25 (m, 3 H), 7.28 - 7.38 (m, 4 H), 7.62 (dd, J = 8.48, 1.90 Hz, 1 H), 7.86 (d, J = 1.90 Hz, 1 H), 7.88−7.95 (m, 2 H), 8.01 (d, J = 8.48 Hz, 1 H), 8.24 (s, 1 H), 8.29 (d, J = 2.19 Hz, 1 H), 8.46 (d, J = 2.19 Hz, 1 H).

*N-(2-Chloro-5-(3-(2-(piperidin-1-yl)ethylamino)quinoxalin-6-yl)-pyridin-3-yl)-4-fluorobenzenesulfonamide (**18k**).* 38.5 mg. Yield 63%. MS (ESI pos. ion) m/z: calc'd for $C_{26}H_{26}ClFN_6O_2S$ 540.2; found 540.8 (M+1). HRMS calc'd for $C_{26}H_{26}ClFN_6O_2S$ (M + H): 541.1579; found 541.1570. ^{1}H NMR (300 MHz, $CDCl_3$) δ 1.85−2.23 (m, 7 H), 2.76 (br s, 2 H), 3.43 (br s, 2 H), 3.66 (br s, 1 H), 4.09 (br s, 2 H), 7.05 (s, 1 H), 7.14−7.22 (m, 2 H), 7.58 (dd, J = 8.62, 2.05 Hz, 1 H), 7.81−7.87 (m, 2 H), 7.89 (d, J = 1.75 Hz, 1 H), 8.02 (d, J = 8.48 Hz, 1 H), 8.34 (d, J = 2.34 Hz, 1 H), 8.40 (s, 1 H), 8.50 (d, J = 2.34 Hz, 1 H).

*N-(2-Chloro-5-(3-(2-morpholinoethylamino)quinoxalin-6-yl)pyridin-3-yl)-4-fluorobenzenesulfonamide (**18l**).* This compound was prepared from **18b** (0.045 g, 0.10 mmol) and 4-(2-aminoethyl)morpholine (0.040 mL, 0.31 mmol) according to the general S_N2' method to afford the title compound (0.043 g, 76%) as an off-white solid. MS (ESI pos. ion) m/z: calc'd for $C_{25}H_{24}ClFN_6O_3S$ 542.1; found 543.0 (M+1). HRMS calc'd for $C_{25}H_{24}ClFN_6O_3S$ (M + H): 543.1372; found 543.1381. ^{1}H NMR (300 MHz, DMSO-d_6) δ 2.55−2.88 (m, 6 H), 3.54−3.77 (m, 6 H), 7.39 (t, J = 8.84 Hz, 2 H), 7.56 (dd, J = 8.48, 2.05 Hz, 1 H), 7.71 (d, J = 1.75 Hz, 2 H), 7.77−7.91 (m, 3 H) 7.97 (d, J = 2.19 Hz, 1 H), 8.37 (s, 1 H), 8.51 (br s, 1 H), 10.28 (br s, 1 H).

*N-(2-Chloro-5-(3-(piperidin-1-yl)quinoxalin-6-yl)pyridin-3-yl)-4-fluorobenzenesulfonamide (**18m**).* 16.2 mg. Yield 32%. MS (ESI pos. ion) m/z: calc'd for $C_{24}H_{21}ClFN_5O_2S$ 497.1; found 498.0 (M+1). HRMS calc'd for $C_{24}H_{21}ClFN_5O_2S$ (M + H): 498.1158; found 498.1170. ^{1}H NMR (300 MHz, $CDCl_3$) δ 1.70−1.86 (m, 6 H), 3.80−3.87 (m, 4 H), 7.04 (s, 1 H), 7.13−7.20 (m, 2 H), 7.53 (dd, J = 8.48, 2.05 Hz, 1 H), 7.81−7.89 (m, 3 H), 7.97 (d, J = 8.48 Hz, 1 H), 8.33 (d, J = 2.34 Hz, 1 H), 8.50 (d, J = 2.34 Hz, 1 H), 8.63 (s, 1 H).

*N-(2-Chloro-5-(3-(4,4-difluoropiperidin-1-yl)quinoxalin-6-yl)pyridin-3-yl)-4-fluorobenzenesulfonamide (**18n**).* 5.2 mg. Yield 9%. MS (ESI pos. ion) m/z: calc'd for $C_{24}H_{19}ClF_3N_5O_2S$ 533.1; found 534.0. HRMS calc'd for $C_{24}H_{19}ClF_3N_5O_2S$ (M + H): 534.0970; found 534.0970. ^{1}H NMR (300 MHz, $CDCl_3$) δ 2.07−2.24 (m, 4 H), 3.97−4.04 (m, 4 H), 7.01 (s, 1 H), 7.12−7.21 (m, 2 H), 7.60 (dd, J = 8.48, 2.05 Hz, 1 H), 7.81−7.91 (m, 3 H), 8.02 (d, J = 8.62 Hz, 1 H), 8.34 (d, J = 2.19 Hz, 1 H), 8.50 (d, J = 2.34 Hz, 1 H), 8.67 (s, 1 H).

*N-(5-(3-((1s,5s)-3-Aza-bicyclo[3.2.2]nonan-3-yl)quinoxalin-6-yl)-2-chloropyridin-3-yl)-4-fluorobenzenesulfonamide (**18o**).* 3.9 mg. Yield 7%. MS (ESI pos. ion) m/z: calc'd for $C_{27}H_{25}ClFN_5O_2S$ 537.1; found 538.0 (M+1). HRMS calc'd for $C_{27}H_{25}ClFN_5O_2S$ (M + H): 538.1470; found 538.1480. ^{1}H NMR (300 MHz, $CDCl_3$) δ 1.76 (t, J = 1.75 Hz, 8 H), 2.29 (br s, 2 H), 4.02 (d, J = 4.09 Hz, 4 H), 7.00 (s, 1 H), 7.12−7.25 (m, 2 H), 7.51 (dd, J = 8.48, 2.05 Hz, 1 H), 7.81−7.91 (m, 3 H), 7.97 (d, J = 8.48 Hz, 1 H), 8.33 (d, J = 2.34 Hz, 1 H), 8.50 (d, J = 2.34 Hz, 1 H), 8.69 (s, 1 H).

*N-(2-Chloro-5-(3-(4-methoxypiperidin-1-yl)quinoxalin-6-yl)pyridin-3-yl)-4-fluorobenzenesulfonamide (**18p**).* 1.8 mg. Yield 3%. MS (ESI pos. ion) m/z: calc'd for $C_{25}H_{23}ClFN_5O_3S$ 527.1; found 528.0 (M+1). HRMS calc'd for $C_{25}H_{23}ClFN_5O_3S$ (M + H): 528.1263; found 528.1271. ^{1}H NMR (300 MHz, $CDCl_3$) δ 1.43 (s, 3 H), 1.77 (br s, 2 H), 1.98−2.10 (m, 2 H), 3.51−3.67 (m, 3 H), 4.10−4.22 (m, 2 H), 7.01 (s, 1 H), 7.12−7.20 (m, 2 H), 7.55 (dd, J = 8.55, 1.97 Hz, 1 H), 7.80−7.89 (m, 3 H), 7.98 (d, J = 8.62 Hz, 1 H), 8.33 (d, J = 2.34 Hz, 1 H), 8.50 (d, J = 2.19 Hz, 1 H), 8.65 (s, 1 H).

*N-(2-Chloro-5-(3-(4-isopropylpiperazin-1-yl)quinoxalin-6-yl)pyridin-3-yl)-4-fluorobenzenesulfonamide (**18q**).* This compound was prepared from **18b** (0.031 g, 0.072 mmol) and 1-isopropylpiperazine (0.026 mL, 0.18 mmol) according to the general S_N2' method to afford the title compound (0.021 g, 54%) as a pale-yellow solid. MS (ESI pos. ion) m/z: calc'd for $C_{26}H_{26}ClFN_6O_2S$ 540.2; found 541.1 (M+1). HRMS calc'd for $C_{26}H_{26}ClFN_6O_2S$ (M + H): 541.1579; found 541.1570. ^{1}H NMR (300 MHz, DMSO-d_6) δ 1.18 (d, J = 6.6 Hz, 6 H),

3.04 (s, 4 H), 3.14−3.23 (m, 1 H), 4.00 (s, 4 H), 7.32 (t, J = 8.8 Hz, 2 H), 7.61 (dd, J = 8.5, 1.9 Hz, 1 H), 7.71 (d, J = 1.8 Hz, 1 H), 7.75−7.86 (m, 2 H), 7.85−7.97 (m, 2 H), 8.26 (s, 1 H), 8.88 (s, 1 H).

*tert-Butyl 3-((7-(6-chloro-5-(4-fluorophenylsulfonamido)pyridin-3-yl)quinoxalin-2-ylamino)methyl)piperidine-1-carboxylate (**18r**).* 21.0 mg. Yield 37%. MS (ESI pos. ion) m/z: calc'd for $C_{30}H_{32}ClFN_6O_4S$ 626.2; found 626.8. ^{1}H NMR (300 MHz, $CDCl_3$) δ 1.38−1.60 (m, 11 H), 1.71 (d, J = 9.21 Hz, 1 H), 1.89−2.10 (m, 3 H), 2.97−3.23 (m, 2 H), 3.56 (br s, 2 H), 3.83 (d, J = 12.86 Hz, 1 H), 7.10−7.24 (m, 3 H), 7.62 (d, J = 8.18 Hz, 1 H), 7.85−7.93 (m, 3 H), 8.04 (d, J = 8.48 Hz, 1 H), 8.27 (d, J = 2.19 Hz, 1 H), 8.45 (d, J = 2.34 Hz, 1 H), 8.50 (br s, 1 H).

*N-(2-Chloro-5-(3-(piperidin-3-ylmethylamino)quinoxalin-6-yl)-pyridin-3-yl)-4-fluorobenzenesulfonamide (**18s**).* A solution of **18r** (5 mg, 8 μmol) in DCM (1 mL) was treated with TFA (1 mL) and stirred at 23 °C for 15 min. The reaction was then concentrated in vacuo. DCM (1 mL) was added and the solvent was evaporated. This procedure was repeated 2 additional times to give the TFA salt of the title compound (5 mg, 100%) as a yellow film. MS (ESI pos. ion) m/z: calc'd for $C_{25}H_{24}ClFN_6O_2S$ 526.1; found 527.0. HRMS calc'd for $C_{25}H_{24}ClFN_6O_2S$ (M + H): 527.1423; found 527.1410. ^{1}H NMR (300 MHz, $CDCl_3$) δ 1.61−1.99 (m, 4 H), 2.17 (d, J = 14.62 Hz, 1 H), 2.64 (t, J = 11.84 Hz, 1 H), 2.71−2.83 (m, 1 H), 3.22 (br s, 1 H), 3.32−3.47 (m, 3 H), 7.02−7.09 (m, 2 H), 7.45 (dd, J = 8.48, 2.05 Hz, 1 H), 7.69−7.77 (m, 3 H), 7.83 (d, J = 8.48 Hz, 1 H), 8.17−8.21 (m, 2 H), 8.37 (d, J = 2.19 Hz, 1 H) (no exchangeable protons were observed because a small amount of MeOD was added for solubility).

■ ASSOCIATED CONTENT

Supporting Information. Biological assays, in vivo study protocols, and X-ray crystallographic data. This material is available free of charge via the Internet at http://pubs.acs.org.

Accession Codes

£The crystal structure of PI3K-gamma in complex with compound **16d** has been deposited in the Protein Data Bank with accession code 3S2A.

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■ ABBREVIATIONS

Ac, acetyl; BID, twice daily; CL, clearance; DCM, dichloromethane; DMF, *N*,*N*-dimethylformamide; DMSO, dimethylsulfoxide; DNA-PK, DNA-dependent serine/threonine protein kinase; dppf, 1, 1′-bis(diphenylphosphino)ferrocene; EC, effective concentration; ED, effective dose; EtOAc, ethyl acetate; HGF, hepatocyte growth factor; HPMC, hydroxypropyl methylcellulose; IV, intravenous; MRT, mean residence time; mTOR, mammalian target of rapamycin; mTORC1, mTOR containing complex 1; MeOH, methanol; NBS, *N*-bromosuccinimide; PD, pharmacodynamic; PI3K, phosphoinositide 3-kinase; PO, orally; PTEN, phosphate and tensin homologue gene; QD, once daily; TBAF, tetrabutylammonium fluoride; TFA, trifluoroacetic acid; Vd,ss, volume of distribution at steady state

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(19) **16d**, **16k** and **18d** were also tested in the Ambit kinase panel at 1 μM. (see Supporting Information).

(20) Experimental details of the PD study can be found in the Supporting Information.

(21) Experimental details of the xenograft study can be found in the Supporting Information.

(22) The doses used in the xenograft study were selected based on the pharmacokinetic and pharmacodynamic profiles of compound **16d** and were estimated to give ≥50% target coverage for up to 24 h.