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Small Molecule Disruptors of the Glucokinase–Glucokinase Regulatory Protein Interaction: 3. Structure–Activity Relationships within the Aryl Carbinol Region of the *N*-Arylsulfonamido-*N*′-arylpiperazine Series

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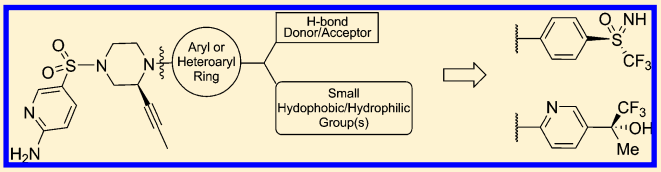
Supporting Information

ABSTRACT: We have recently reported a novel approach to increase cytosolic glucokinase (GK) levels through the binding of a small molecule to its endogenous inhibitor, glucokinase regulatory protein (GKRP). These initial investigations culminated in the identification of 2-(4-((2*S*)-4-((6-amino-3-pyridinyl)sulfonyl)-2-(1-propyn-1-yl)-1-piperazinyl)phenyl)-1,1,1,3,3,3-hexafluoro-2-propanol (**1**, AMG-3969), a compound that effectively enhanced GK translocation and reduced blood glucose levels in diabetic animals. Herein we report the results of our expanded SAR investigations that focused on modifications to the aryl carbinol group of this series. Guided by the X-ray cocrystal structure of compound **1** bound to hGKRP, we identified several potent GK–GKRP disruptors bearing a diverse set of functionalities in the aryl carbinol region. Among them, sulfoximine and pyridinyl derivatives **24** and **29** possessed excellent potency as well as favorable PK properties. When dosed orally in *db*/*db* mice, both compounds significantly lowered fed blood glucose levels (up to 58%).



INTRODUCTION

Diabetes affects ∼370 million people worldwide, and the health expenditures related to diabetes have been estimated to be over 471 billion dollars per year.[1] Current treatments for diabetes include patient education (lifestyle changes), insulin injections, and antidiabetic drugs (to control blood glucose levels). Despite many years of efforts to effectively manage diabetes, there is still a great unmet medical need for orally available, safe, and effective drugs for the treatment of diabetes.[2] Allosteric activation of glucokinase (GK) by small molecules has recently been targeted by several pharmaceutical companies for the potential treatment of diabetes.[3] Even though this approach has had some initial success (glucose lowering effect in animals and in phase I clinical trials), undesirable side effects (e.g., hypoglycemia) and loss of efficacy in longer term phase II clinical trials have hindered the advancement of this class of antidiabetic agents.[4] Other strategies to minimize hypoglycemic risk have included designing GK activators that are exclusively taken up by the liver.[5] These molecules are substrates for organic anion transporters and therefore result in selective activation of hepatic GK.

Glucokinase regulatory protein (GKRP) plays a key role in controlling GK levels in hepatocytes by releasing GK from the nucleus when the blood glucose level is high and by binding to GK and sequestering it inside the nucleus when the glucose level is low.[6] By disrupting the GK–GKRP interaction, we postulated that more unbound GK would be available to regulate glucose levels, and since the kinetic parameters of GK (V_{max} and $S_{0.5}$) would not be altered by this mechanism, the risk of hypoglycemia would be reduced.

In our previous publications, we described the identification of a series of piperazine-based small molecule GK–GKRP disruptors.[7,8] These investigations led to the development of metabolically stable GK–GKRP disruptors that were potent and efficacious, culminating in the identification of compound **1** (AMG-3969) and its close analogue **2**.[9] Both compounds were potent (IC_{50} < 10 nM) in the biochemical assay (AlphaScreen), which measured the compound's abilities to disrupt the GK–GKRP interaction.[10] They were also effective (EC_{50} ≈ 100–200 nM) in the functional mouse hepatocyte assay that measured the translocation of GK from the nucleus to the cytoplasm.[11] Compounds **1** and **2** also had good in vivo pharmacokinetic (PK) properties in rats (*F* of 75% and 82%, respectively) and were found to significantly lower blood glucose levels in *db*/*db* mice.

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In our preliminary SAR studies, the sulfonamide (region A in Figure 1) and the central core piperazine (region B in Figure 1) areas were extensively investigated; however, the SAR around the phenyl carbinol (region C in Figure 1) was very limited.[8,9]

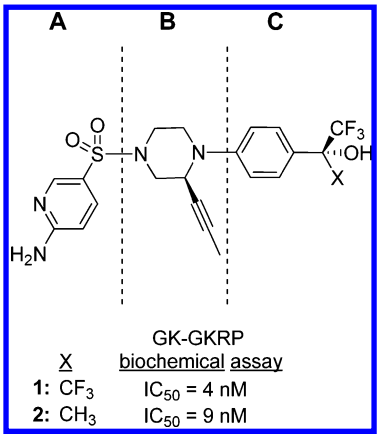

Figure 1. Region representation of compounds **1** and **2**.

In these initial investigations, the C region was restricted to only phenyl-bis-trifluoromethyl carbinol (e.g., **1**) and phenylmethyltrifluoromethyl carbinol (e.g., **2**), which had comparable potencies. Therefore, we wanted to explore the SAR around the C region in more detail. Toward that end, we first examined the X-ray structure of compound **1** bound to hGKRP (Figure 2).[9] To focus better in this area, only the residues in proximity to the C region of compound **1** are highlighted in the structures of Figure 2.[12] As can be seen from each of the four views, the phenyl C-ring of compound **1** sits deep within the binding pocket and occupies a hydrophobic area of the protein. This C-ring is sandwiched between Ala521 on the top and Val28 on the bottom with Glu32 and Tyr24 residues on either side (top and bottom views). The carbinol oxygen forms a strong hydrogen bond with Arg525, while the two trifluoromethyl groups occupy two channels, one projecting toward Phe526, Met522, and His504 and the other projecting toward Lys33 and Ser34 (side and end views).

To further expand the SAR in this region and increase the chemical diversity within this series, we set out with three main goals (as illustrated in general structure **3** in Figure 3): (1) to probe the channels occupied by the two trifluoromethyl groups

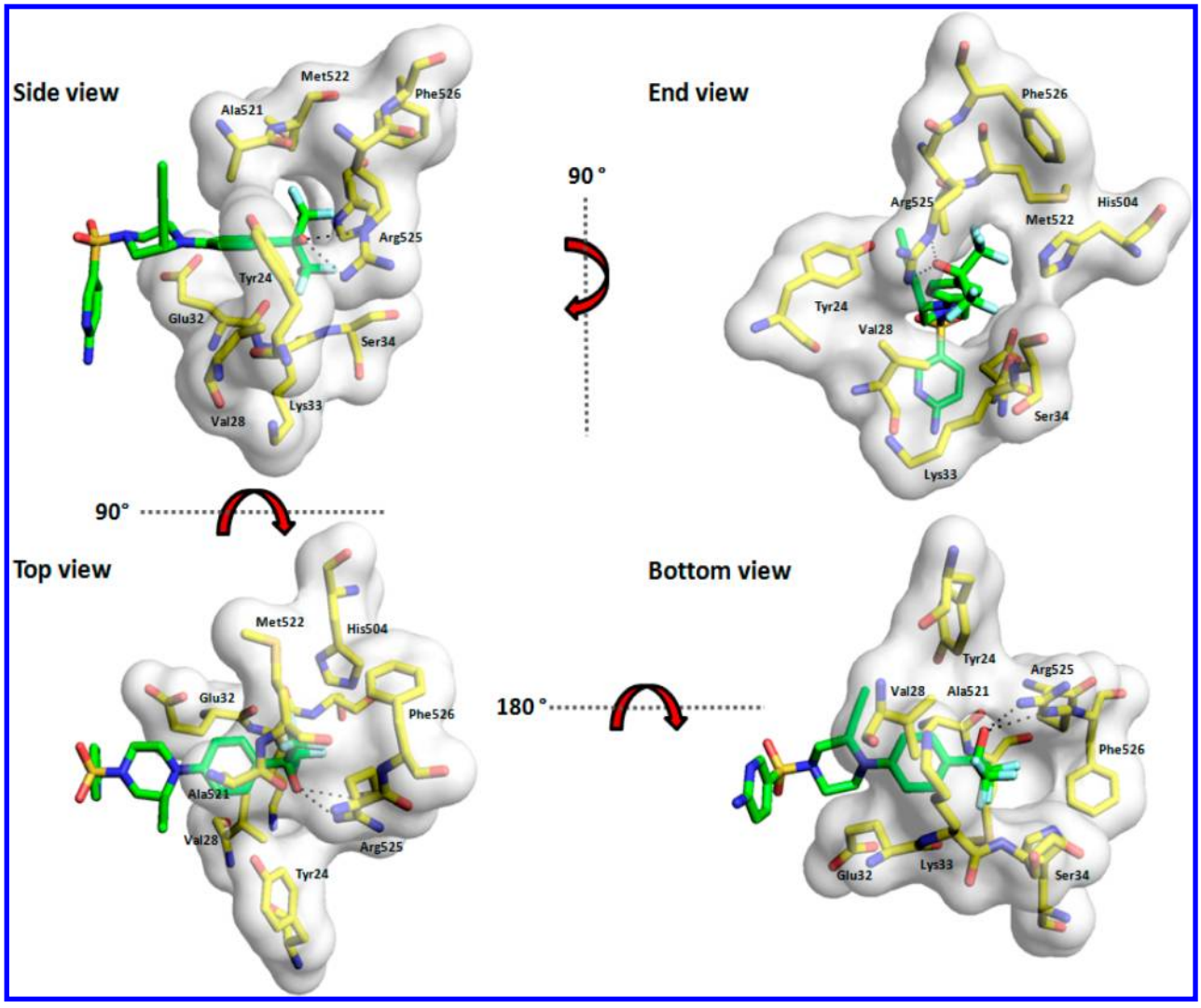

Figure 2. X-ray cocrystal structure of compound **1** bound to full-length hGKRP. Note that only the residues within ~5.5 Å of the C-region of compound **1** are illustrated.

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with small hydrophobic and hydrophilic substituents; (2) to evaluate non-carbinol functional groups that could engage in with Arg525; (3) to examine the effect of various five- and six-membered heterocyclic replacements to the phenyl C-ring.

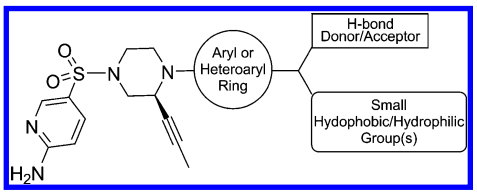


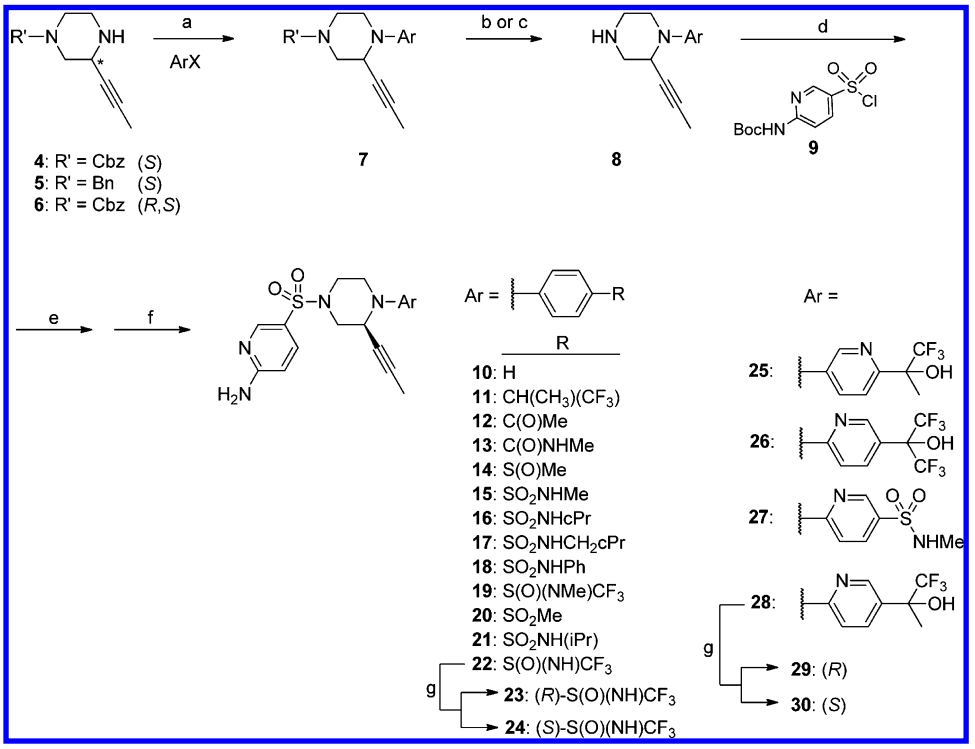
Figure 3. Area of SAR in a general structure **3**.

CHEMISTRY

Because of the structural diversity of this series, several synthetic approaches were necessary to prepare the desired analogues. These methods are summarized in Schemes 1−5. The most general method used to prepare C-ring analogues of compounds **1** and **2** is outlined in Scheme 1. *N*-Carboxybenzyl (Cbz) and *N*-benzyl (Bn) protected 2-alkynylpiperazine[13] **4**, **5**, or **6** were coupled with the appropriate aryl halides (ArX) utilizing palladium catalyzed amination conditions[14] to afford arylpiperazine intermediates **7**. The requisite aryl halides employed in the coupling reactions were either commercially available or prepared as described in the Supporting Information. For the majority of the cases, deprotection of the Cbz group was achieved by treatment with triflic acid in trifluoroacetic acid. Alternatively, the Cbz group was removed by treating with sulfuric acid in chloroform. In the case of *N*-benzyl intermediates **7**, the compounds were treated with 1-chloroethyl chlorocarbonate followed by heating in methanol to provide the corresponding deprotected intermediates **8**.[15] *N*-Arylpiperazines **8** were reacted with *tert*-butyl (5-(chlorosulfonyl)pyridin-2-yl)carbamate (**9**)[13] and triethylamine or diisopropylethylamine in dichloromethane. Subsequent cleavage of the *tert*-butyloxycarbonyl (Boc) group with trifluoroacetic acid afforded the target sulfonamides **10**−**19** and **25**−**28**. The methylsulfone and isopropylsulfonamide derivatives (**20** and **21**, respectively) were obtained by separation of the corresponding racemic intermediates by chiral supercritical fluid chromatography (SFC) (these derivatives were prepared from the racemic 2-alkynylpiperazine, **6**). Compounds **22** and **28** were also subjected to chiral SFC purification to afford the pure diastereomers **23** and **24** as well as **29** and **30**, respectively.[16]

Scheme 2 outlines the synthesis of compounds that required further modifications to the C-ring substituent after they were coupled with the 2-alkynylpiperazine (**4** or **5**). The synthesis of (*S*)-cyclopropylsulfoximine **47** started by coupling 2-alkynylpiperazine **5** with 1-bromo-4-(cyclopropylsulfanyl)benzene[13] to obtain intermediate **31**. The coupled product was then debenzylated and reacted with sulfonyl chloride **9** to give **41**, which was oxidized with hydrogen peroxide in acetic acid to afford sulfoxide **42**. Treatment of compound **42** with sodium azide and sulfuric acid in chloroform provided the targeted sulfoximine **47**.[17] The corresponding methylsulfoximine derivative, **48**, was prepared by the analogous method from 1-bromo-4-(methylsulfinyl)benzene[13] and 2-alkynylpiperazine

Scheme 1. Synthesis of C-Ring Analogues 10−30[a]



[a]Reagents and conditions: (a) RuPhos/RuPhos first generation catalyst, RuPhos/$Pd_2(dba)_3$, DavePhos/$Pd_2(dba)_3$, X-Phos/$Pd_2(dba)_3$, JohnPhos/$Pd_2(dba)_3$, or BINAP/$Pd(OAc)_2$, NaO-*t*-Bu, 1,4-dioxane or toluene; (b) TfOH, TFA or conc H_2SO_4, $CHCl_3$; (c) (i) 1-chloroethyl chlorocarbonate, K_2CO_3, DCM; (ii) MeOH, reflux; (d) TEA or DIPEA, DCM; (e) TFA, DCM; (f) chiral SFC separation (for compounds starting from intermediate **6**); (g) chiral SFC separation.[16]

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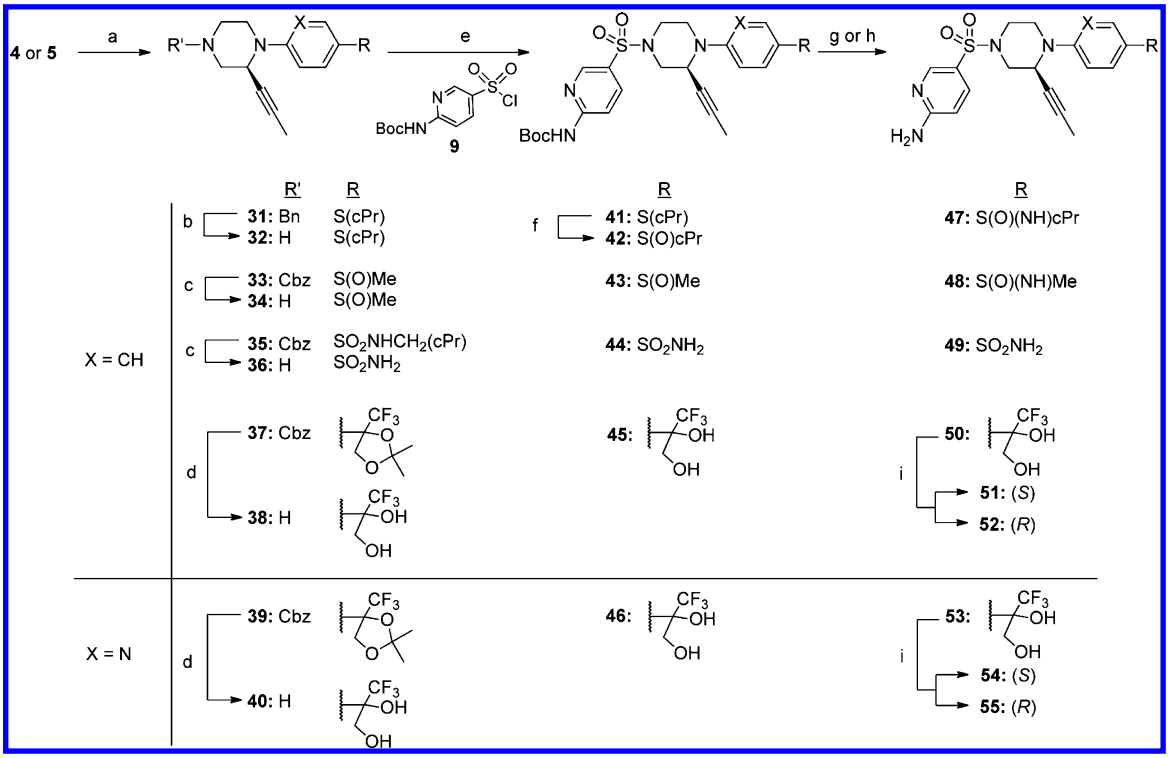
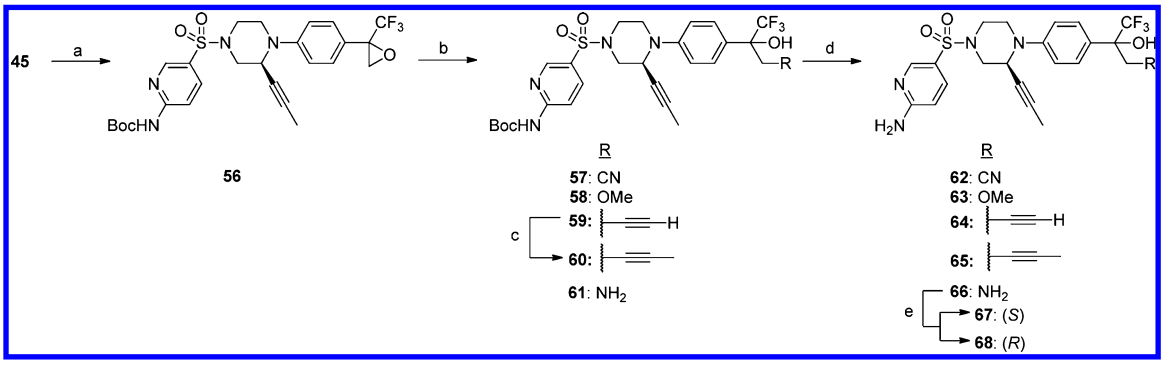
Scheme 2. Synthesis of C-ring Analogues 47−55[a]



[a]Reagents and conditions: (a) ArX, RuPhos/RuPhos first generation catalyst, DavePhos/$Pd_2(dba)_3$, or JohnPhos/$Pd_2(dba)_3$, NaO-*t*-Bu, 1,4-dioxane or toluene; (b) (i) 1-chloroethyl chlorocarbonate, K_2CO_3, DCM; (ii) MeOH reflux; (c) H_2SO_4, $CHCl_3$; (d) TfOH, TFA; (e) TEA or DIPEA, DCM; (f) H_2O_2, AcOH; (g) TFA, DCM; (h) NaN_3, H_2SO_4, $CHCl_3$; (i) chiral SFC purification (the absolute stereochemistries of compounds **51** and **52** were established based on X-ray cocrystal structures with hGKRP, and the stereochemistries of compounds **54** and **55** were arbitrarily assigned).

Scheme 3. Synthesis of Trifluoromethyl Carbinol Analogues 62−68[a]



[a]Reagents and conditions: (a) TsCl, TEA, DCM. (b) For **57**: KCN, water, DMF. For **58**: NaOMe, DMF, MeOH. For **59**: (i) *n*-BuLi, ethynyltrimethylsilane, THF; (ii) K_2CO_3, MeOH. For **61**: NH_4OH, DMF. (c) *n*-BuLi, MeI, THF; (d) TFA, DCM; (e) chiral SFC purification (the absolute stereochemistries of compounds **67** and **68** were arbitrarily assigned).

4. For the compound with the primary sulfonamide substituent, **49**, the synthesis began with the coupling of 2-alkynylpiperazine **4** with 4-bromo-*N*-(cyclopropylmethyl)benzenesulfonamide[13] to obtain **35**. Under strongly acidic conditions, the Cbz group and the cyclopropylmethyl group were removed to give **36**. Subsequent sulfonamide formation with *tert*-butyl (5-(chlorosulfonyl)pyridin-2-yl)carbamate (**9**) followed by deprotection of the Boc group afforded the primary sulfonamide **49**. The compounds bearing trifluoromethyldiol substituents with either a phenyl or pyridine C-ring (**50** or **53**, respectively) were constructed in a similar manner to each other. Coupling reactions of 2-alkynylpiperazine **4** with the appropriate aryl bromides[13] containing the protected diols afforded **37** for the phenyl derivative and **39** for the pyridine analogue. Removing the Cbz groups with triflic acid in trifluoroacetic acid also served to deprotect the acetonide to provide intermediates **38** and **40**, respectively. Subsequent sulfonamide formation and Boc removal afforded the diols **50** and **53**, respectively. The diastereomeric diols were resolved by chiral SFC purification to obtain the pure epimers. From diol **50**, epimers **51** and **52** were obtained in >98% de, and the absolute stereochemistry was assigned from X-ray crystallographic data from the ligand−protein (hGKRP) complex. Similarly, epimers **54** and **55** were separated from diol **53**. However, in this case, the stereo-

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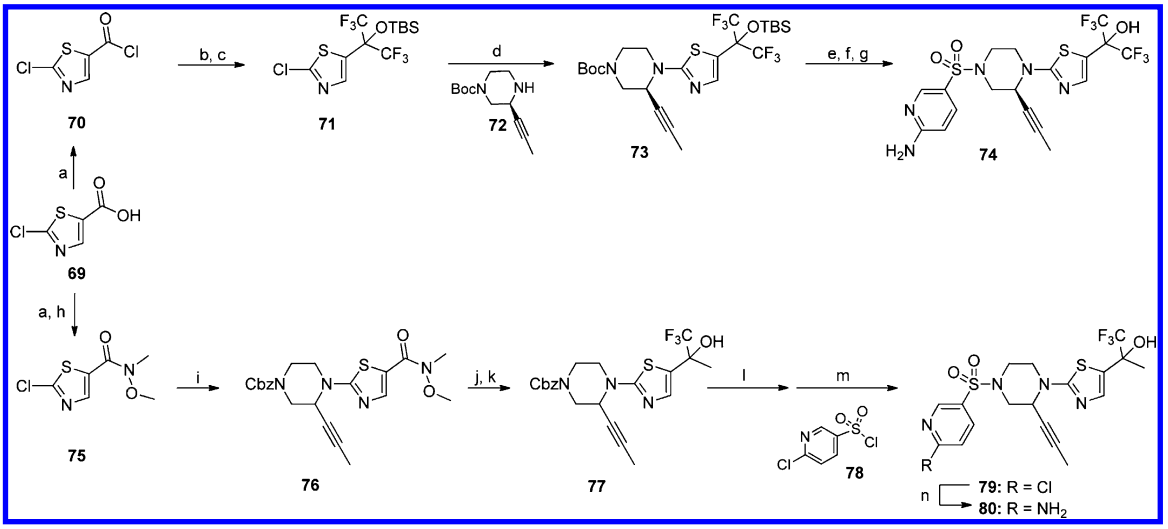
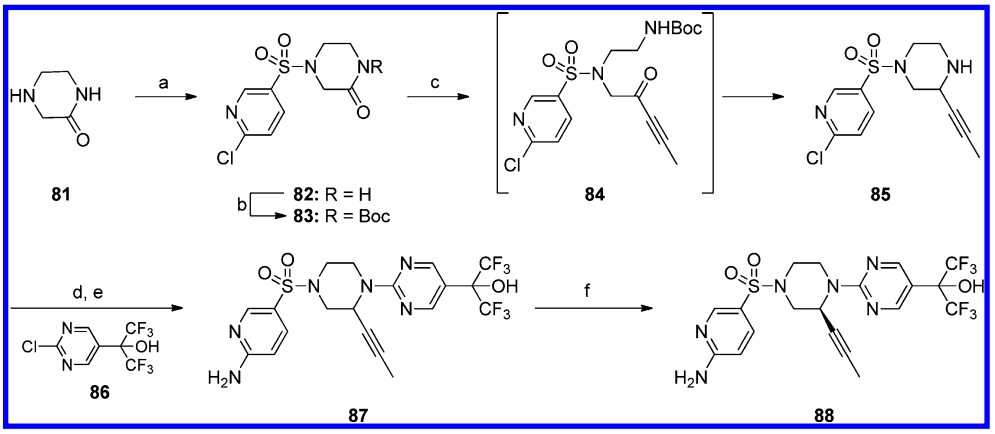
Scheme 4. Synthesis of Thiazole C-Ring Analogues 74 and 80[a]



[a]Reagents and conditions: (a) oxalyl chloride, cat. DMF, DCM; (b) $TMSCF_3$, TMAF, DME; (c) TBSOTf, DIPEA, DCM; (d) RuPhos, RuPhos first generation catalyst, NaO-*t*-Bu, 1,4-dioxane; (e) TFA, DCM; (f) **9**, TEA, DCM; (g) HF–pyridine, ACN, TFA; (h) *N*,*O*-dimethylhydroxylamine–HCl, DIPEA, DCM; (i) **6**, DIPEA, DMF; (j) MeMgBr, THF; (k) $TMSCF_3$, TMAF, THF; (l) TfOH, TFA; (m) TEA, DCM; (n) NH_4OH, EtOH.

Scheme 5. Synthesis of Pyrimidine 88[a]



[a]Reagents and conditions: (a) **78**, TEA, DCM; (b) $(Boc)_2O$, TEA, DMAP, DCM; (c) (i) bromo(1-propy-1-yl)magnesium, THF; (ii) TFA, DCM; (iii) $NaBH(OAc)_3$; (d) **86**, DIPEA, NMP; (e) (i) NH_4OH, EtOH; (ii) $NaBH_4$, MeOH;[20] (f) chiral SFC purification.

chemistries at the carbinol stereocenters of **54** and **55** were assigned based on their order of elution compared to that of the phenyl analogues (**51** and **52**) and the absolute stereochemistries were not unambiguously determined.

Compounds with additional modifications to the trifluoromethyl carbinol center were synthesized by opening the epoxide of intermediate **56** (prepared from the diol intermediate **45**) with various nucleophiles (Scheme 3). For example, treatment of **56** with potassium cyanide, sodium methoxide, lithiated ethynyltrimethylsilane (followed by removal of TMS group), or ammonium hydroxide gave intermediates **57**, **58**, **59**, or **61**, respectively. The methylalkyne derivative **60** was obtained by methylation of compound **59** with methyl iodide. Removal of the Boc groups of compounds **57**–**61** with trifluoroacetic acid yielded the final products **62**–**66**. The diastereomeric amine compound **66** was subjected to chiral SFC chromatography to provide diastereomers **67** and **68** with >98% diastereomeric excess. The absolute stereochemistries at the carbinol carbons **67** and **68** were arbitrarily assigned.

Both thiazole C-ring analogues **74** and **80** were prepared from commercially available 2-chloro-1,3-thiazole-5-carboxylic acid (**69**), and their syntheses are outlined in Scheme 4. For the bis-trifluoromethyl carbinol derivative, the trifluoromethyl group was installed first by converting acid **69** to acid chloride **70** followed by treatment with Ruppert's reagent (trimethylsilyltrifluoromethane) and tetramethylammonium fluoride.[18] The resulting hydroxyl group was then protected as the *tert*-butyldimethylsilyl ether to afford **71**. Subsequent amination with Boc-protected piperazine **72**[13] afforded **73**, which was carried through the sequence of Boc removal, sulfonamide formation (with sulfonyl chloride **9**), and a one-pot removal of both the *tert*-butyldimethylsilyl and Boc groups to obtain thiazole **74**. For the monotrifluoromethyl derivative **80**, acid **69** was converted to Weinreb amide **75** that was then coupled with piperazine **6** via nucleophilic substitution condition to form **76**. The amide moiety of **76** was converted to the monotrifluoromethyl carbinol via ketone formation with methylmagnesium bromide followed by trifluoromethylation to obtain **77**.[18] Deprotection of the Cbz group followed by

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sulfonamide formation with sulfonyl chloride **78** provided chloropyridine sulfonamide **79**. The requisite aminopyridine **80** was obtained by treatment of **79** with ammonium hydroxide in ethanol.

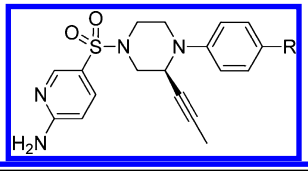
The synthesis of the final analogue, pyrimidine **88**, is illustrated in Scheme 5. Commercially available ketopiperazine **81** was reacted with sulfonyl chloride **78**[19] to form the corresponding sulfonamide, **82**. The amide NH of **82** was protected as the *tert*-butyl carbamate to give **83**. Treatment of **83** with the bromo(1-propy-1-yl)magnesium initially gave the opened methyl ketone **84**. Removal of the Boc group, cyclization to the corresponding imine, and subsequent reduction with sodium triacetoxyborohydride afforded the piperazine **85**. Compound **85** was coupled with chloropyrimidine **86**[13] via nucleophilic aromatic substitution, and the resulting chloropyridine was converted to aminopyridine **87**. Chiral SFC purification of the racemate **87** gave the pure enantiomers. The absolute stereochemistry of the carbon bearing the propargyl group of the more active enantiomer (**88**) was assigned to be (*S*) configuration, since we have established from our previous SAR studies that compounds with the (*S*) configuration at this center were significantly more potent than their corresponding (*R*) epimers.[8,9]

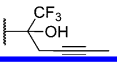
RESULTS AND DISCUSSION

The compounds prepared in this study were tested in a hGK–hGKRP binding assay (IC_{50}, μM) and a GK translocation assay (EC_{50}, μM) in mouse hepatocytes.[10,11] In addition to the two primary assays, the analogues were evaluated for their metabolic stability in rat liver microsomes (RLM) as measured by the intrinsic clearance, CL_{int} (μL min^{-1} mg^{-1}).[21,22] The current structure–activity relationship (SAR) investigation focused on two general areas of the molecule: the type of substituent on the 4 position of the C-ring, which we refer to as the "tail region" (Tables 1 and 2), and the nature of the aromatic C-ring itself (Table 3).

Table 1 summarizes the results for compounds with either small hydrophilic or hydrophobic groups in the tail region of the molecule or with no substituent at all. The importance of the carbinol functionality is clearly illustrated by compounds **10** and **11**. No activity was observed for compound **10** where no tail group was present, and only micromolar activity was obtained for compound **11** where the alcohol moiety was absent, despite the presence of the hydrophobic trifluoromethyl/methyl groups. Since removal of the trifluoromethyl carbinol functionality resulted in more than a 100-fold decrease in potency (presumably due to the loss of the hydrogen bond to Arg525), we maintained the trifluoromethyl carbinol functionality and explored additional substitutions on the methyl group in the remaining analogues reported in Table 1. These additional substituents were selected to probe the two narrow channels observed in this area of the GKRP protein. The upper channel projects toward residues Phe526, Met522, and His504, and the lower channel is flanked by residues Ser34, Lys33, and Val28 (see Figure 1, end view). An additional hydroxyl group adjacent to the tertiary carbinol was well tolerated, as shown by the diols **51** and **52**. Isomer **51** also exhibited improved in vitro stability with liver microsomal clearance of <14 μL min^{-1} mg^{-1}. Interestingly, the amino analogues (**67** and **68**) were less potent compared to compound **2**, suggesting that basic groups are not as tolerated in the region of the protein. In the X-ray structure of diol **51** bound to GKRP (Figure 4A, orange),[23] the primary hydroxyl group forms hydrogen bonds with the

Table 1. Biochemical and Cellular Assay Results with Rat Liver Microsomal (RLM) Stability of Compounds with Modifications to the Trifluoromethyl Carbinol Center



Cmpd Number	R	hGK-hGKRP AlphaScreen IC_{50} (μM)[a]	Mouse Translocation EC_{50} (μM)[b]	RLM CL_{int} (μL/min/mg)[c]
1		0.004	0.202	42
2		0.009	0.120	39
10		25.0	NA	>399
11		1.05	1.56	>399
51		0.028	0.098	<14
52		0.006	0.089	53
67[d]		0.374	5.67	46
68[d]		0.047	0.537	56
62		0.028	0.219	57
63		0.016	0.202	93
64		0.006	0.130	59
65		0.005	0.137	115

[a]AlphaScreen data reported as an average ($n \geq 3$). Compounds with activities of >12.5 μM, $n = 1$. For all others, standard deviations are reported in the Supporting Information. [b]Mouse hepatocyte translocation data reported as an average ($n \geq 3$). Standard deviations ($n \geq 3$) are reported in the Supporting Information. [c]Average of two experimental values. [d]The stereochemistry at the carbinol carbon was arbitrarily assigned.

imidazole N of His504 (N···OH) and the NH of Arg525 (NH···O); however, the NH_2 groups of **67** and **68** would be partially charged and unable to serve as hydrogen-bond acceptors for His504. Both cyano (**62**) and methoxy (**63**) groups were well tolerated when compared to compound **2**, although the cell potency suffered slightly. When small hydrophobic groups such as ethynal and propargyl were introduced next to the carbinol group (**64** and **65**), the analogues showed potent activities similar to those of compounds **1** and **2**. In the X-ray structure of propargyl derivative **65** bound to GKRP (Figure 4B, purple),[23] the configuration of the trifluorocarbinol was very similar to that of compound **1** (Figure 4B, light-green lines). The alcohol moiety forms a hydrogen bond to Arg525, and the propargyl group projects up into the narrow channel near Phe526, Met522, and His504. In addition, one of the hydrogens of the propargyl

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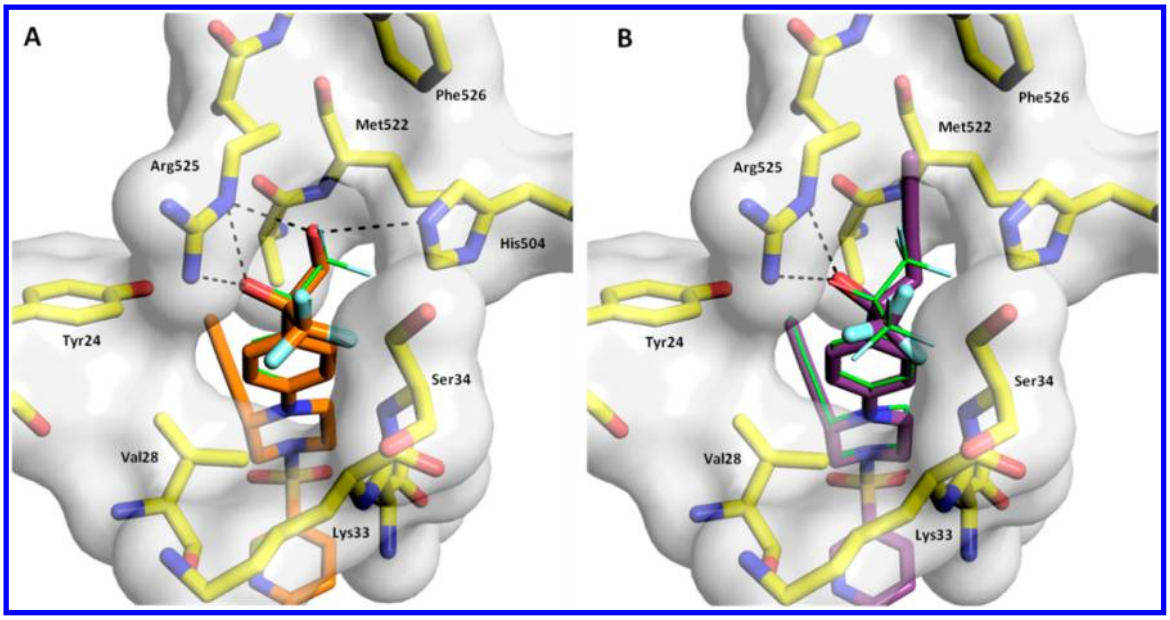

Figure 4. (A) X-ray cocrystal structure of compound **51** (orange sticks) bound to hGKRP determined at 2.4 Å resolution. (B) X-ray cocrystal structure of compound **65** (purple sticks) bound to hGKRP determined at 2.8 Å resolution. The X-ray crystal structure of compound **1** (light-green lines) is superimposed in both (A) and (B), and the gray surface indicates the van der Waals surface area.

methyl group engages in an edge-to-face interaction with Phe526. Overall, the data reveal that small groups capable of forming either hydrogen-bond interactions or hydrophobic interactions with protein residues near the tail region were tolerated and the trifluoropropanediol group (compounds **51** and **52**) offered the best potency and metabolic stability profile from this initial set of analogues.

Table 2 summarizes our efforts to identify suitable bioisosteric replacements for the trifluoromethyl carbinol functionality. As discussed previously, the C-ring carbinol engages in a key hydrogen-bonding interaction with Arg525. Since the basic Arg525 side chain normally exists as the protonated form at neutral pH, the C-ring carbinol most likely plays the role of a hydrogen-bond acceptor through the lone pairs of electrons on the oxygen atom. Therefore, we explored the effect of other oxygen containing groups. Simple carbonyl compounds such as ketone **12** and amide **13** were approximately 100-fold less potent in the biochemical assay when compared to compounds **1** and **2**. It is likely that these carbonyl compounds lacked the three-dimensional feature of our lead compounds that was important in filling the hydrophobic pocket adjacent to Arg525. For this reason, we turned our attention to sulfur-based oxygen functionalities as alternative tail group moieties. Although the simple sulfoxide and sulfone derivatives (**14** and **20**, respectively) were only weak to moderate disruptors, we were encouraged by the potent activity of sulfonamide **49** (binding IC_{50} = 61 nM and cellular EC_{50} = 37 nM). The potent activity of compound **49** was somewhat surprising, since the sulfonamide tail region lacked any hydrophobic groups that had been shown earlier to be essential for the carbinol series. Therefore, we examined derivatives with N-substitution on the sulfonamide, postulating that potency might be enhanced by adding hydrophobic groups in this region to gain additional van der Waals contacts to either the upper or lower channel of the GKRP protein. Methylsulfonamide **15** showed a modest improvement in the biochemical assay; however, cellular potency was not improved. The X-ray crystal structure of compound **15** bound to the GKRP protein (Figure 5A) showed that the two oxygens on C-ring sulfonamide were engaged in hydrogen-bonding interactions with the Arg525 and the sulfonamide methyl group extended toward the lower Ser34/Lys33/Val28 channel; however, there seemed to be no additional interaction between the sulfonamide NH and the protein. Unfortunately, attempts to gain additional activity by extending further into the lower channel by increasing the size of the substituent on the nitrogen of the sulfonamide were unsuccessful. As the size of the hydrophobic group increased (from methyl to cyclopropyl, isopropyl, methylcyclopropyl, and phenyl; compounds **16**, **21**, **17**, and **18**, respectively), activity decreased. These sulfonamide analogues also exhibited reduced microsomal stability in rat liver microsomes.

Lastly, sulfoximines were examined, as they offer certain unique properties compared to sulfones and sulfonamides.[24,25] For example, sulfoximines have been successfully employed as alcohol isosteres.[26] In addition, while being isosteric with sulfones, they also introduced a mildly basic nitrogen that could be alkylated to give an added substituent. Since the sulfur center is chiral, we postulated that the sulfoximine could also resemble the chiral trifluoromethyl carbinol group in compound **2**. Both methyl- and cyclopropylsulfoximines **48** and **47** showed modest biochemical activities, with the latter also exhibiting good cellular activity (EC_{50} = 0.208 μM). The trifluoromethylsulfoximine **22** was approximately 7 times more potent than the methyl analogue **48** in the biochemical assay. The two diastereomers of sulfoximine **22** (**23** and **24**) showed 10-fold differential activities, with the (*S*)-isomer **24** being the most potent sulfoximine analogue. Notably, the sulfoximines also exhibited marked microsomal stability overall; however, when the nitrogen of sulfoximine **24** was methylated (compound **19**), both the potency and the microsomal stability suffered. In this phase of the investigation we were able to replace the carbinol function with either a sulfonamide or a sulfoximine group while maintaining good potency and stability. Furthermore, our design hypothesis was supported by the X-ray structures of sulfonamide **15** and sulfoximine **24**

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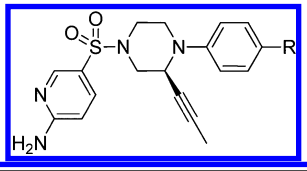
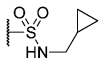
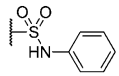
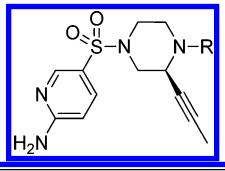
Table 2. Biochemical and Cellular Assay Results with Rat Liver Microsomal (RLM) Stability of Compounds with Carbinol Replacements

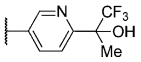
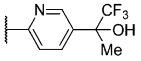
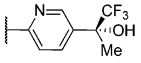
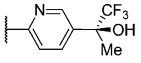
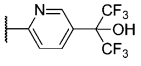
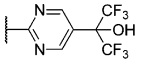
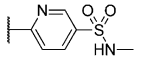
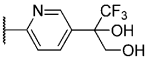
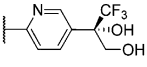
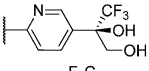
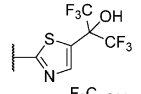
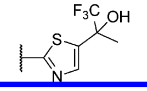


Cmpd Number	R	hGK-hGKRP AlphaScreen IC_{50} (μM)[a]	Mouse Translocation EC_{50} (μM)[b]	RLM CL_{int} (μL/min/mg)[c]
12		1.09	>12.5	249
13		3.07	4.05	41
14		3.36	>12.5	17
20		0.401	0.936	34
49		0.061	0.037	30
15		0.035	0.097	55
16		0.101	0.254	136
21		0.235	0.178	129
17		0.320	0.271	213
18		1.60	>12.5	>399
48		0.310	0.704	<14
47		0.256	0.208	23
22		0.045	0.567	38
23		0.182	2.22	47
24		0.017	0.388	21
19		0.266	2.06	257

[a]AlphaScreen data reported as an average ($n \geq 3$). Standard deviations are reported in the Supporting Information. [b]Mouse hepatocyte translocation data reported as an average ($n \geq 2$). Compounds with activities of >12.5 μM, $n = 1$. For all others, standard deviations ($n \geq 2$) are reported in the Supporting Information. [c]Average of two experimental values.

Table 3. Biochemical and Cellular Assay Results with Rat Liver Microsomal (RLM) Stability of Compounds with Heterocycle C-Rings



Cmpd Number	R	hGK-hGKRP AlphaScreen IC_{50} (μM)[a]	Mouse Translocation EC_{50} (μM)[b]	RLM CL_{int} (μL/min/mg)[c]
25		0.069	1.45	31
28		0.018	0.294	23
29		0.013	0.160	29
30		0.013	0.382	38
26		0.010	0.421	42
88		0.010	2.20	28
27		0.068	0.381	42
53		0.024	0.145	18
54[d]		0.013	0.349	31
55[d]		0.070	0.155	<14
74		0.081	>12.5	34
80[e]		0.049	>12.5	87

[a]AlphaScreen data reported as an average ($n \geq 3$). Standard deviations are reported in the Supporting Information. [b]Mouse hepatocyte translocation data reported as an average ($n \geq 3$). Compounds with activities of >12.5 μM, $n = 1$. For all others, standard deviations ($n \geq 3$) are reported in the Supporting Information. [c]Average of two experimental values. [d]The stereochemistry at carbinol carbon was arbitrarily assigned. [e]This compound is a mixture of four stereoisomers.

bound to the GKRP protein (Figure 5A and Figure 5B).[23] Both structures show a bifurcated interaction between Arg525 and either the O═S═O or the O═S═N functional group in sulfonamide **15** or sulfoximine **24**, respectively. In the case of sulfoximine **24** (Figure 5B, light-blue), the trifluoromethyl group projects toward Ser34 and resides in a position similar to that of the lower trifluoromethyl group of compound **1** (Figure 5B, light-green).

The SAR results for compounds bearing a heterocyclic C-ring are summarized in Table 3. Compared to the monotrifluoromethyl carbinol, **2**, the 2-pyridinyl derivative **25** was 7-fold less potent in the biochemical assay and 10-fold less potent in the cellular assay; however, the 3-pyridyl analogue **28** showed comparable biochemical potency. The two diastereomers of pyridine **28** (**29** and **30**) were equipotent, with diastereomer **29** showing similar cellular potency as that of compound **2**. When the 3-pyridyl C-ring was combined with the bis-trifluoromethyl carbinol, the cellular potency dropped 2-fold compared to compound **1**, even though potency of the binding assay was preserved (compound **26**). A further drop in

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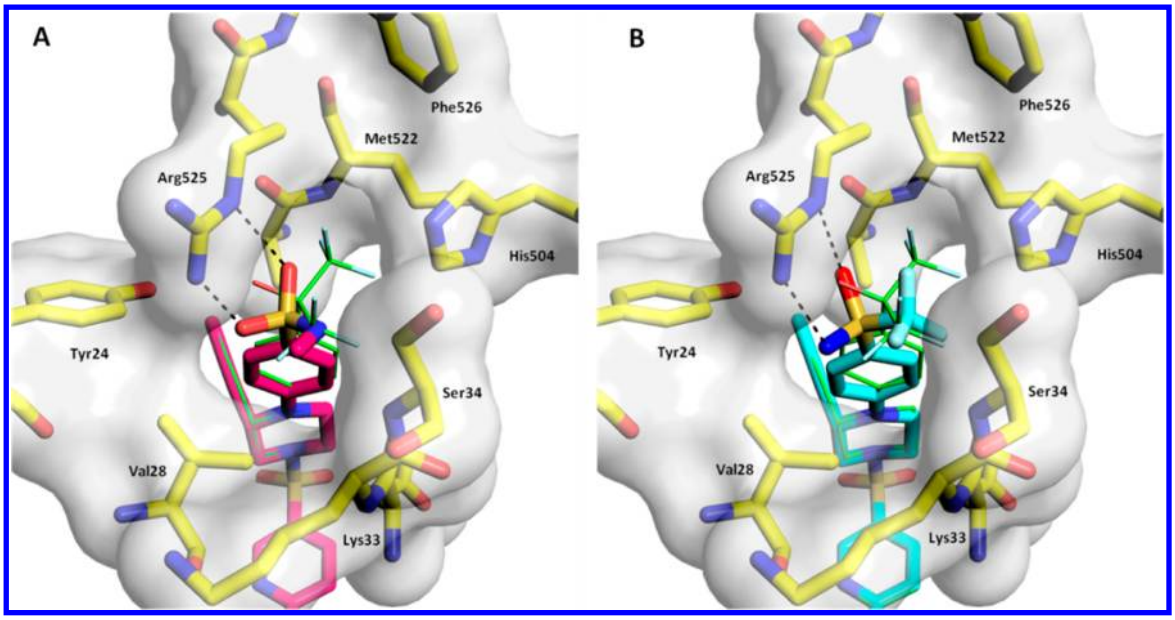

Figure 5. (A) X-ray cocrystal structure of compound **15** (magenta sticks) bound to hGKRP determined at 2.4 Å resolution. (B) X-ray cocrystal structure of compound **24** (light-blue sticks) bound to hGKRP determined at 2.58 Å resolution. The X-ray crystal structure of compound **1** (light-green lines) is superimposed in both (A) and (B), and the gray surface indicates the van der Waals surface area.

cellular potency (~5-fold) was observed when an additional nitrogen was added in the C-ring (i.e., pyrimidine **88**). In general, analogues with the bis-trifluoromethyl carbinol tail group showed a steeper shift in biochemical to cellular potency when compared to the monotrifluoromethyl carbinol series. We hypothesize that the higher logP of the former might be the underlying factor. For example, the bis-trifluoromethyl analogue **26** has a higher clogP value (2.8) and larger cell shift (42-fold) than the corresponding methyltrifluoromethyl analogue, **28** (clogP = 2.2 and a cell shift of 16-fold).[27] This trend was also observed between compounds **1** and **2**, where compound **1** (clogP = 3.7) showed a 49-fold cell shift and compound **2** (clogP = 3.2) had only a 12-fold cell shift.

With the promising results obtained with 3-pyridyl C-ring derivatives **26** and **28**–**30** (good cellular potencies and good metabolic stabilities in RLM), we combined the pyridyl C-ring with two of the better tail groups identified previously to give methylsulfonamide pyridine **27** and trifluoropropanediol pyridine **53**. The two diastereomers of diol **53** were also separated and evaluated (**54** and **55**). Overall, addition of the pyridine nitrogen was well tolerated; however, small losses in potencies were observed when the 3-pyridine C-ring replaced the phenyl ring (i.e., methylsulfonamides (**27** vs **15**) and trifluoropropanediols (**54** and **55** vs **51** and **52**). Finally, both of the two five-membered C-ring analogues that were examined showed reduced biochemical and cellular potencies (thiazoles **74** and **80**). It is likely that the orientation of the carbinol group in the five-membered heterocycle is not optimal to provide as strong of a hydrogen-bonding interaction with Arg525, compared with compounds **1** and **2**.

The SAR studies described above, which focused on the C-ring modifications to compounds **1** and **2**, resulted in the identification of several potent GK–GKRP disruptors with good metabolic stability in rat liver microsomes. To evaluate these derivatives further, several of the potent analogues were selected for additional pharmacokinetic studies. A variety of structural differences were represented in the selected compounds (i.e., trifluoropropanediol **51**, sulfonamide **49**, methylsulfonamide **15**, trifluoromethylsulfoximine **24**, and pyridinetrifluoromethyl carbinol **29**). The results are shown in Table 4, and the pharmacokinetic profile of compound **1** is included for comparison. Despite their relatively low in vitro microsomal clearance values, sulfonamides **49** and **15** showed high in vivo clearance in rats (2.8 and 3.7 L kg^{-1} h^{-1}, respectively) as well as low oral bioavailabilities (F of 5% and 1.5%, respectively). Trifluoropropanediol **51** also gave low exposures when administered orally (AUC = 7.1 μM·h) and showed relatively high clearance (1.2 L kg^{-1} h^{-1}) considering its stability in RLM (<14 μL min^{-1} mg^{-1}). On the other hand, sulfoximine **24** and pyridine **29** were found to have favorable pharmacokinetic profiles in rats with low clearance values, good oral bioavailabilities, and high exposures. Therefore, sulfoximine **24** and pyridine **29** were also tested in mouse pharmacokinetic experiments to determine if these derivatives would be suitable for efficacy studies in mice. The clearance of sulfoximine **24** was very low (0.025 L kg^{-1} h^{-1}) resulting in even higher exposures in mice than in rats (362 vs 25 μM·h at 10 mg/kg, respectively). The pharmacokinetic profile of pyridine **29** was found to be similar in both mice and rats.

Given their potent cellular activities and their favorable pharmacokinetic profiles, we evaluated the efficacy of sulfoximine **24** and pyridine **29** in *db*/*db* mice. The results of these studies are shown in Figure 6A and Figure 6B, respectively. Both compounds were found to lower blood glucose levels when orally administered to *db*/*db* mice. As shown in Figure 6A, compound **24** caused statistically significant reductions (up to 58%) at all doses (10, 30, 100 mg/kg) at both 3 and 6 h time points. Like pyridine **24**, sulfoximine **29** (Figure 6B) also caused significant decreases in fed blood glucose levels at both the early (3 and 6 h) time points. With the exception of the 100 mg/kg dose of **24**, glucose levels had returned to normal 24 h postdose.

CONCLUSION

We have described the synthesis and biological evaluation of a series of *N*-arylsulfonamido-*N*′-arylpiperazine GK–GKRP

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Table 4. Pharmacokinetic Properties of Selected Compounds

	in vivo rat PK[a]				in vivo mouse PK[d]					
	iv[b]		po[c]		iv[b]		po[c]			
compd	CL (L kg^{-1} h^{-1})	$t_{1/2}$ (h)	F (%)	AUC (μM·h)	CL (L kg^{-1} h^{-1})	$t_{1/2}$ (h)	F (%)	AUC (μM·h)	C_{max} (μM)	T_{max} (h)
1	0.75	3.6	75	20.0	0.11	6.7	40	64.8	5.82	5.0
49	2.82	1.6	5	0.26						
15	6.68	4.1	1.5	0.03						
51	1.26	3.9	48	7.1						
24	0.42	2.8	51	25.3	0.025	3.1	53	362	46.6	1.0
29	0.35	3.2	47	28.4	0.28	2.4	47	37.0	9.71	0.25

[a]Pharmacokinetic parameters following administration in male Sprague−Dawley rats: n = 3 for each time point, 3 animals per study. [b]Dosed at 2 mg/kg as a solution in DMSO. [c]Dosed orally at 10 mg/kg (for **49**, 1% Tween 80, 2% hydroxypropyl methylcellulose (HPMC); all others, 1% Tween 80, 2% HPMC, pH 2.2 with methanesulfonic acid (MSA). [d]Pharmacokinetic parameters following administration in male C57BL/6 mice: n = 3 for each time point, 9 animals per study.

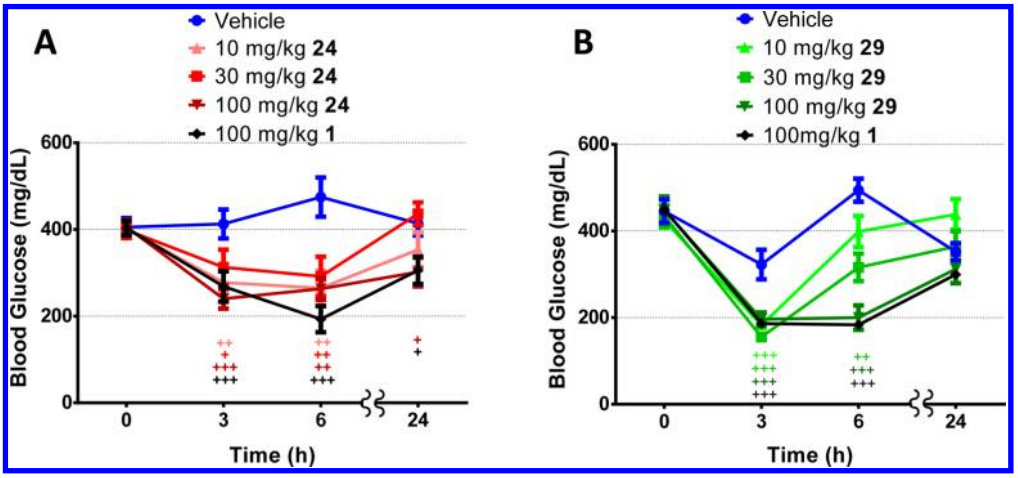

Figure 6. (A) Blood glucose measurements after oral administration of **24** (10, 30, 100, mg/kg) to *db*/*db* mice. (B) Blood glucose measurements after oral administration of **29** (10, 30, 100, mg/kg) to *db*/*db* mice. The statistical significance of the blood glucose measurements were based on comparison to the individual vehicle control groups ($n \geq 6$, * = $p < 0.05$, ** = $p < 0.01$, *** = $p < 0.001$) calculated by ANOVA.

disruptors with modifications to the phenyl carbinol C-ring. Guided by structure-based design utilizing the X-ray cocrystal structure of compound **1** bound to hGKRP, we explored three main aspects of the C-ring SAR including exploration of the two channels accessed by the terminal C-ring substituent (tail region), assessment of various carbinol isosteres designed to engage Arg525, and evaluation of heterocyclic replacements to the phenyl C-ring. Through these investigations we were able to identify several diverse and potent C-region analogues, including diols (**51** and **52**), alkynes (**64** and **65**), sulfonamides (**49** and **15**), sulfoximine (**24**), and pyridines (**29**, **30**, **54**, and **55**). Among these derivatives, compound **24** (trifluoromethyl-sulfoximine as the carbinol bioisostere) and compound **29** (pyridine replacement for the phenyl C-ring) were found to possess favorable in vivo pharmacokinetic properties in rodents (F_{oral} = 47−53% in both rat and mouse), and when dosed orally in *db*/*db* mice, they produced significant reductions in blood glucose levels (~40−58% 6 h postdose at 100 mg/kg). These novel compounds will be evaluated further to assess their potential as therapeutic antidiabetic agents.

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. 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). NMR spectra were determined with a Bruker 300 MHz or DRX 400 MHz spectrometer. Chemical shifts are reported in parts per million (ppm, δ units). All final compounds were purified to >95% purity unless otherwise noted as determined by LC/MS obtained on an Agilent 1100 or 1200 spectrometer using the following methods: [A] Agilent SB-C18 column (50 mm × 3.0 mm, 2.5 μm) 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] Waters XBridge C18 column (50 mm × 3.0 mm, 3 μm) 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 spectrometry (MS) data were obtained at the same time of the purity determination on the LC/MS instrument using ES ionization mode (positive).

General Amination Method. To a reaction vessel were added benzyl (3*S*)-3-(1-propyn-1-yl)-1-piperazinecarboxylate (**4**), (3*S*)-1-benzyl-3-(1-propyn-1-yl)piperazine (**5**), or benzyl 3-(1-propyn-1-yl)-1-piperazinecarboxylate (**6**),[13] an aryl halide (1.1−1.5 equiv), ligand (RuPhos, DavePhos, X-Phos JohnPhos, or BINAP, 5−10 mol %), Pd source (RuPhos first generation precatalyst, $Pd_2(dba)_3$, or $Pd(OAc)_2$, 10 mol %), NaO-*t*-Bu (2−3 equiv), and 1,4-dioxane or toluene. The mixture was deoxygenated by bubbling argon gas through it and was then heated at 80−100 °C until the starting material was consumed. The mixture was allowed to cool to room temperature and partitioned between EtOAc and water. The aqueous phase was extracted with EtOAc. The combined organic phases were washed with saturated aqueous NaCl, dried over Na_2SO_4 or $MgSO_4$, filtered, and

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concentrated. The material was purified by silica gel column chromatography.

General *N*-Carboxylbenzyl (Cbz) Deprotection: Method A. A Cbz-protected amine was dissolved in TFA. Trifluoromethanesulfonic acid (3 equiv) was added slowly at room temperature, and the mixture was stirred for 2−5 min. The reaction mixture was concentrated and subjected to the next reaction directly or quenched as follows: Solid $NaHCO_3$ was added to the reaction mixture slowly followed by aqueous saturated $NaHCO_3$. The aqueous phase was extracted with EtOAc, and the combined organic phases were washed with water and aqueous saturated NaCl. The organic phase was dried over Na_2SO_4 or $MgSO_4$, filtered, and concentrated. The crude product was used directly in the next step or purified by silica gel column chromatography.

General *N*-Carboxylbenzyl (Cbz) Deprotection: Method B. To a reaction vessel was added a Cbz-protected amine and $CHCl_3$, and the mixture was cooled to 0 °C. Excess concentrated H_2SO_4 was added, and the mixture was stirred at room temperature until the starting material was consumed. The reaction mixture was basified by the addition of 10% NaOH and then extracted with EtOAc. The organic extract was washed with water, aqueous saturated NaCl, then dried over anhydrous Na_2SO_4 and filtered. The filtrate was concentrated under reduced pressure. The crude product was used directly in the next step or purified by silica gel column chromatography.

General *N*-Benzyl (Bn) Deprotection Method. To a reaction vessel were added a Bn-protected amine, 1-chloroethyl chlorocarbonate (5 equiv), K_2CO_3 (3 equiv), and DCM. The mixture was stirred at room temperature until the starting material was consumed. The solid was removed by filtration, and the filtrate was concentrated. MeOH was added to the residue, and the mixture was heated at reflux for 2 h. The mixture was allowed to cool to room temperature and concentrated. The crude product was used directly in the next step or purified by silica gel column chromatography.

General Sulfonamide Formation Method. To a reaction vessel was added a deprotected piperazine, Et_3N, DIPEA or pyridine (3−10 equiv), and DCM. After the mixture was stirred for 5 min, appropriate sulfonyl chloride (1.1 equiv) was added. The mixture was stirred at room temperature until the starting material was consumed. The mixture was concentrated in vacuo and used directly in the next deprotection step or purified by silica gel chromatography.

General *N*-*tert*-Butyl Carbamate (Boc) Deprotection Method. To a reaction vessel was added a Boc protected aminopyridine sulfonamide, TFA (20−50 equiv), and DCM. The mixture was stirred at room temperature until the reaction was complete. The reaction mixture was concentrated and then neutralized by the addition of saturated aqueous $NaHCO_3$. The aqueous phase was extracted with EtOAc, and the combined organic phases were washed with saturated aqueous $NaHCO_3$, water, and saturated aqueous NaCl. The organic phase was dried over Na_2SO_4 or $MgSO_4$, filtered, and concentrated. The crude product was purified by silica gel column chromatography unless otherwise noted.

5-(((3*S*)-4-Phenyl-3-(1-propyn-1-yl)-1-piperazinyl)sulfonyl)-2-pyridinamine (10). Benzyl (3*S*)-3-(1-propyn-1-yl)-1-piperazinecarboxylate (**4**) (0.250 g, 0.96 mmol) and bromobenzene (0.304 g, 1.93 mmol) were coupled according to the general amination method using 2′-(diphenylphosphino)-*N*,*N*-dimethyl-[1,1′-biphenyl]-2-amine (DavePhos) (0.038 g, 0.097 mmol), $Pd_2(dba)_3$ (0.044 g, 0.048 mmol), and toluene to give benzyl (3*S*)-4-phenyl-3-(1-propyn-1-yl)-1-piperazinecarboxylate (0.200 g, 46%) as a yellow solid. MS (ESI positive ion) *m*/*z*: calcd for $C_{21}H_{22}N_2O_2$, 334.168; found, 335.1 (M + 1). ^{1}H NMR (400 MHz, DMSO-d_6) δ 7.52−7.33 (m, 5 H), 7.29 (t, *J* = 7.9 Hz, 2 H), 7.03 (d, *J* = 8.3 Hz, 2 H), 6.89 (t, *J* = 7.2 Hz, 1 H), 5.30−5.06 (m, 2 H), 4.68 (br s, 1 H), 4.14 (d, *J* = 12.2 Hz, 2 H), 3.04 (br s, 2 H), 1.69 (d, *J* = 1.7 Hz, 3 H). Two protons were obscured by the water peak.

Benzyl (3*S*)-4-phenyl-3-(1-propyn-1-yl)-1-piperazinecarboxylate (0.250 g, 0.749 mmol) was deprotected according to the general Cbz-deprotection method B to give (2*S*)-1-phenyl-2-(1-propyn-1-yl)piperazine (0.150 g, crude). The crude product was used in the next step without any purification. MS (ESI positive ion) *m*/*z*: calcd for $C_{13}H_{16}N_2$, 200.131; found, 201.1 (M + 1).

(2*S*)-1-Phenyl-2-(1-propyn-1-yl)piperazine (0.150 g, 0.75 mmol) was treated with *tert*-butyl (5-(chlorosulfonyl)-2-pyridinyl)carbamate (**9**) (0.260 g, 0.9 mmol) according to the general sulfonamide formation method using Et_3N (0.2 mL, 1.5 mmol) to give *tert*-butyl (5-(((3*S*)-4-phenyl-3-(1-propyn-1-yl)-1-piperazinyl)sulfonyl)-2-pyridinyl)carbamate (0.120 g, 35%) as a white solid. MS (ESI positive ion) *m*/*z*: calcd for $C_{23}H_{28}N_4O_4S$, 456.183; found, 457.1 (M + 1).

tert-Butyl (5-(((3*S*)-4-phenyl-3-(1-propyn-1-yl)-1-piperazinyl)-sulfonyl)-2-pyridinyl)carbamate (0.120 g, 0.305 mmol) was deprotected according to the general Boc-deprotection method to give 5-(((3*S*)-4-phenyl-3-(1-propyn-1-yl)-1-piperazinyl)sulfonyl)-2-pyridinamine (**10**) (0.020 g, 19%) as a white solid. MS (ESI positive ion) *m*/*z*: calcd for $C_{18}H_{20}N_4O_2S$, 356.131; found, 356.9 (M + 1). ^{1}H NMR (400 MHz, DMSO-d_6) δ 8.23 (d, *J* = 2.5 Hz, 1 H), 7.64 (dd, *J* = 2.5, 8.8 Hz, 1 H), 7.26−7.18 (m, 2 H), 7.03 (s, 2 H), 6.94 (d, *J* = 8.0 Hz, 2 H), 6.83 (t, *J* = 7.3 Hz, 1 H), 6.54 (d, *J* = 8.8 Hz, 1 H), 4.72 (br s, 1 H), 3.63−3.53 (m, 2 H), 3.47−3.41 (m, 1 H), 3.13−3.03 (m, *J* = 8.8, 11.9 Hz, 1 H), 2.56 (dd, *J* = 3.3, 11.3 Hz, 1 H), 2.42−2.34 (m, 1 H), 1.73 (d, *J* = 1.9 Hz, 3 H).

5-(((3*S*)-3-(1-Propyn-1-yl)-4-(4-(2,2,2-trifluoro-1-methylethyl)phenyl)-1-piperazinyl)sulfonyl)-2-pyridinamine (11). Benzyl (3*S*)-3-(1-propyn-1-yl)-1-piperazinecarboxylate (**4**) (0.520 g, 2.01 mmol) and 1-bromo-4-(2,2,2-trifluoro-1-methylethyl)-benzene[13] (0.470 g, 1.86 mmol) were coupled according to the general amination method using RuPhos first generation precatalyst (65 mg, 0.080 mmol) and 1,4-dioxane to afford benzyl (3*S*)-3-(1-propyn-1-yl)-4-(4-(2,2,2-trifluoro-1-methylethyl)phenyl)-1-piperazinecarboxylate (0.38 g, 47%) as a clear oil. MS (ESI positive ion) *m*/*z*: calcd for $C_{24}H_{25}F_3N_2O_2$, 430.187; found, 431.3 (M + 1). ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.27−7.44 (m, 5 H), 7.22 (d, *J* = 7.43 Hz, 2 H), 6.94 (d, *J* = 7.82 Hz, 2 H), 5.05−5.33 (m, 2 H), 4.10−4.40 (m, 3 H), 3.02−3.43 (m, 5 H), 1.68 (br s, 3 H), 1.47 (d, *J* = 6.85 Hz, 3 H).

Benzyl (3*S*)-3-(1-propyn-1-yl)-4-(4-(2,2,2-trifluoro-1-methylethyl)-phenyl)-1-piperazinecarboxylate (0.38 g, 0.88 mmol) was deprotected according to the general Cbz-deprotection method A to afford (2*S*)-2-(1-propyn-1-yl)-1-(4-(2,2,2-trifluoro-1-methylethyl)phenyl)piperazine as a crude product. This material was used for the next reaction without further purification.

(2*S*)-2-(1-Propyn-1-yl)-1-(4-(2,2,2-trifluoro-1-methylethyl)phenyl)-piperazine (0.26 g, crude) was treated with *tert*-butyl (5-(chlorosulfonyl)-2-pyridinyl)carbamate (**9**) (0.300 g, 1.03 mmol) according to the general sulfonamide formation method using Et_3N (0.600 mL, 4.30 mmol) aand then deprotected by the general Boc-deprotection method to afford 5-(((3*S*)-3-(1-propyn-1-yl)-4-(4-(2,2,2-trifluoro-1-methylethyl)phenyl)-1-piperazinyl)sulfonyl)-2-pyridinamine (**11**) (0.315 g, 79% over three steps) as a light-yellow foam. MS (ESI positive ion) *m*/*z*: calcd for $C_{21}H_{23}F_3N_4O_2S$, 452.149; found, 453.2 (M + 1). ^{1}H NMR (400 MHz, $CDCl_3$) δ 8.49 (br s, 1 H), 7.78 (d, *J* = 9.19 Hz, 1 H), 7.21 (d, *J* = 7.24 Hz, 2 H), 6.91 (d, *J* = 7.04 Hz, 2 H), 6.54 (d, *J* = 7.24 Hz, 1 H), 5.03 (br s, 2 H), 4.37 (br s, 1 H), 3.72 (t, *J* = 11.83 Hz, 2 H), 3.27−3.43 (m, 3 H), 2.86 (d, *J* = 11.35 Hz, 1 H), 2.71 (t, *J* = 10.56 Hz, 1 H), 1.78 (br s, 3 H), 1.46 (d, *J* = 6.06 Hz, 3 H).

1-(4-((2*S*)-4-((6-Amino-3-pyridinyl)sulfonyl)-2-(1-propyn-1-yl)-1-piperazinyl)phenyl)ethanone (12). (3*S*)-1-Benzyl-3-(1-propyn-1-yl)piperazine (**5**) (0.500 g, 2.52 mmol) and 1-(4-bromophenyl)-ethanone (0.540 g, 2.52 mmol) were coupled according to the general amination method using RuPhos (0.059 g, 0.13 mmol), RuPhos first generation precatalyst (0.061 g, 0.075 mmol), and 1,4-dioxane to afford 1-(4-((2*S*)-4-benzyl-2-(1-propyn-1-yl)-1-piperazinyl)phenyl)-ethanone (0.120 g, 16%) as an off-white solid. MS (ESI positive ion) *m*/*z*: calcd for $C_{22}H_{24}N_2O$, 332.189; found, 333.1 (M + 1). ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.91 (d, *J* = 8.8 Hz, 2 H), 7.44−7.27 (m, 5 H), 6.94 (d, *J* = 8.8 Hz, 2 H), 4.53 (s, 1 H), 3.74 (d, *J* = 13.4 Hz, 1 H), 3.58−3.51 (m, 2 H), 3.36−3.67 (m, 1 H), 3.04−2.98 (m, 2 H), 2.53 (s, 3 H), 2.38−2.33 (m, 2 H), 1.80 (d, *J* = 2.2 Hz, 3 H).

1-(4-((2*S*)-4-Benzyl-2-(1-propyn-1-yl)-1-piperazinyl)phenyl)-ethanone (0.130 g, 0.39 mmol) was deprotected according to the general Bn-deprotection method to afford 1-(4-((2*S*)-2-(1-propyn-1-

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yl)-1-piperazinyl)phenyl)ethanone (0.090 g). This material was used without further purification. MS (ESI positive ion) *m/z*: calcd for $C_{15}H_{18}N_2O$, 242.142; found, 243.0 (M + 1).

1-(4-((2*S*)-2-(1-Propyn-1-yl)-1-piperazinyl)phenyl)ethanone (0.090 g, 0.37 mmol) was treated with *tert*-butyl (5-(chlorosulfonyl)-2-pyridinyl)carbamate (**9**) (0.217 g, 0.74 mmol) according to the general sulfonamide formation method using pyridine (0.15 mL, 1.85 mmol) to afford *tert*-butyl (5-(((3*S*)-4-(4-acetylphenyl)-3-(1-propyn-1-yl)-1-piperazinyl)sulfonyl)-2-pyridinyl)carbamate (0.075 g, 41%) as a white solid. MS (ESI positive ion) *m/z*: calcd for $C_{25}H_{30}N_4O_5S$, 498.194; found, 399.0 (M − Boc + 1). ^{1}H NMR (400 MHz, $CDCl_3$) δ 8.68 (d, J = 2.4 Hz, 1 H), 8.15 (d, J = 8.9 Hz, 1 H), 8.05−7.99 (m, 2 H), 7.90 (d, J = 8.8 Hz, 2 H), 6.92 (d, J = 8.8 Hz, 2 H), 4.57 (s, 1 H), 3.86−3.80 (m, 2 H), 3.54−3.56 (m, 1 H), 3.41−3.40 (m, 1 H), 2.82 (dd, J = 11.1, 3.4 Hz, 1 H), 2.68−2.66 (m, 1 H), 2.53 (s, 3 H), 1.78 (d, J = 2.1 Hz, 3 H), 1.55 (s, 9 H).

tert-Butyl (5-(((3*S*)-4-(4-acetylphenyl)-3-(1-propyn-1-yl)-1-piperazinyl)sulfonyl)-2-pyridinyl)carbamate (0.075 g, 0.15 mmol) was deprotected according to the general Boc-deprotection method to afford 1-(4-((2*S*)-4-((6-amino-3-pyridinyl)sulfonyl)-2-(1-propyn-1-yl)-1-piperazinyl)phenyl)ethanone (**12**) (0.030 g, 50%) as an off-white solid. MS (ESI positive ion) *m/z*: calcd for $C_{20}H_{22}N_4O_3S$, 398.141; found, 399.0 (M + 1). ^{1}H NMR (400 MHz, $CDCl_3$) δ 8.23 (d, J = 2.5 Hz, 1 H), 7.83−7.81 (m, 2 H), 7.64−7.61 (m, 1 H), 7.03−6.99 (m, 4 H), 6.53 (d, J = 8.9 Hz, 1 H), 4.96 (s, 1 H), 3.74 (d, J = 11.6 Hz, 1 H), 3.64−3.59 (m, 2 H), 3.12−3.11 (m, 1 H), 2.54 (d, J = 3.6 Hz, 1 H), 2.49 (s, 3 H), 2.39−2.37 (m, 1 H), 1.74 (d, J = 1.9 Hz, 3 H).

4-((2*S*)-4-((6-Amino-3-pyridinyl)sulfonyl)-2-(1-propyn-1-yl)-1-piperazinyl)-*N*-methylbenzamide (13). Benzyl (3*S*)-3-(1-propyn-1-yl)-1-piperazinecarboxylate (**4**) (0.484 g, 1.87 mmol) and 4-bromo-*N*-methylbenzamide[13] (0.40 g, 1.9 mmol) were coupled according to the general amination method using $Pd_2(dba)_3$ (0.034 g, 0.037 mmol), X-Phos (0.026 g, 0.056 mmol), and 1,4-dioxane to afford benzyl (3*S*)-4-(4-(methylcarbamoyl)phenyl)-3-(1-propyn-1-yl)-1-piperazinecarboxylate (0.22 g, 30%) as a white solid. MS (ESI positive ion) *m/z*: calcd for $C_{23}H_{25}N_3O_3$, 391.190; found, 392.1 (M + 1).

Benzyl (3*S*)-4-(4-(methylcarbamoyl)phenyl)-3-(1-propyn-1-yl)-1-piperazinecarboxylate (0.21 g, 0.54 mmol) was deprotected according to the general Cbz-deprotection method B to obtain *N*-methyl-4-((2*S*)-2-(1-propyn-1-yl)-1-piperazinyl)benzamide (0.13 g, crude) as a yellow solid. The crude product was used in the next step without purification. MS (ESI positive ion) *m/z*: calcd for $C_{15}H_{19}N_3O$: 257.153; found, 258.1 (M + 1).

N-Methyl-4-((2*S*)-2-(1-propyn-1-yl)-1-piperazinyl)benzamide (0.13 g, 0.505 mmol) was treated with *tert*-butyl (5-(chlorosulfonyl)-2-pyridinyl)carbamate (**9**) (0.147 g, 0.505 mmol) according to the general sulfonamide formation method using Et_3N (0.105 mL, 0.757 mmol) to afford *tert*-butyl (5-(((3*S*)-4-(4-(methylcarbamoyl)phenyl)-3-(1-propyn-1-yl)-1-piperazinyl)sulfonyl)-2-pyridinyl)carbamate as an off-white solid. The crude product was used in the next step without purification. MS (ESI positive ion) *m/z*: calcd for $C_{25}H_{31}N_5O_5S$, 513.205; found, 514.1 (M + 1).

tert-Butyl (5-(((3*S*)-4-(4-(methylcarbamoyl)phenyl)-3-(1-propyn-1-yl)-1-piperazinyl)sulfonyl)-2-pyridinyl)carbamate (0.25 g,0.49 mmol) was deprotected according to the general Boc-deprotection method. The crude product obtained was purified by preparative HPLC (Zorbax-C18 XDB, 20−60% (1:1 acetonitrile/MeOH) in (0.1% TFA in water) in 6 min, 20 mL/min) to afford 4-((2*S*)-4-((6-amino-3-pyridinyl)sulfonyl)-2-(1-propyn-1-yl)-1-piperazinyl)-*N*-methylbenzamide (**13**) (0.060 g, 30%) as an off-white solid. MS (ESI positive ion) *m/z*: calcd for $C_{20}H_{23}N_5O_3S$, 413.152; found, 414.1 (M + 1). ^{1}H NMR (400 MHz, CD_3OD) δ 8.23 (d, J = 2 Hz, 1 H), 7.86−7.84 (dd, J = 2.4,9.2 Hz, 1 H), 7.62 (d, J = 8.8 Hz, 2 H), 6.91 (d, J = 8.8 Hz, 2 H), 6.78 (d, J = 8.8 Hz, 1 H), 4.64 (s, 1 H), 3.69 (d, J = 11.2 Hz, 2 H), 3.49 (d, J = 12.8 Hz 1 H), 2.78−2.74 (m, 5 H), 2.62−2.56 (m, 1 H), 1.65 (s, 3 H).

5-(((3*S*)-4-(4-(Methylsulfinyl)phenyl)-3-(1-propyn-1-yl)-1-piperazinyl)sulfonyl)-2-pyridinamine (14). Benzyl (3*S*)-3-(1-propyn-1-yl)-1-piperazinecarboxylate (**4**) (0.50 g, 1.9 mmol) and 1-bromo-4-(methylsulfinyl)benzene[13] (0.63 g, 2.9 mmol) were coupled according to the general amination method using $Pd_2(dba)_3$ (0.089 g, 0.096 mmol), DavePhos (0.037 g, 0.096 mmol), and toluene to afford benzyl (3*S*)-4-(4-(methylsulfinyl)phenyl)-3-(1-propyn-1-yl)-1-piperazinecarboxylate (0.75 g, 65%) as a pale-brown gummy solid. MS (ESI positive ion) *m/z*: calcd for $C_{22}H_{24}N_2O_3S$, 396.151; found, 397.0 (M + 1).

Benzyl (3*S*)-4-(4-(methylsulfinyl)phenyl)-3-(1-propyn-1-yl)-1-piperazinecarboxylate (0.75 g, 1.9 mmol) was deprotected according to the general Cbz-deprotection method B to give (2*S*)-1-(4-(methylsulfinyl)phenyl)-2-(1-propyn-1-yl)piperazine (0.40 g, 80%) as a brown gummy liquid. MS (ESI positive ion) *m/z*: calcd for $C_{14}H_{18}N_2OS$, 262.114; found, 263.1 (M + 1).

(2*S*)-1-(4-(Methylsulfinyl)phenyl)-2-(1-propyn-1-yl)piperazine (0.40 g, 1.5 mmol) was treated with *tert*-butyl (5-(chlorosulfonyl)-2-pyridinyl)carbamate (**9**) (0.67 g, 2.3 mmol) according to the general sulfonamide formation method using Et_3N (0.60 mL, 3.8 mmol) to give *tert*-butyl (5-(((3*S*)-4-(4-(methylsulfinyl)phenyl)-3-(1-propyn-1-yl)-1-piperazinyl)sulfonyl)-2-pyridinyl)carbamate (0.35 g, 44%) as a brown solid. MS (ESI positive ion) *m/z*: calcd for $C_{24}H_{30}N_4O_5S_2$, 518.166; found, 519.3 (M + 1). ^{1}H NMR (300 MHz, $CDCl_3$) δ 8.66 (s, 1 H), 8.13 (d, J = 6.6 Hz, 1 H), 8.03 (dd, J = 6.6, 1.5 Hz, 1 H), 7.65 (s, 1 H), 7.56 (d, J = 6.6 Hz, 2 H), 7.03 (d, J = 6.0 Hz, 2 H), 4.47 (s, 1 H), 3.84−3.77 (m, 2 H), 3.43−3.39 77 (m, 3 H), 2.84 (dd, J = 8.4, 2.7 Hz, 1 H), 2.70 (s, 3 H), 1.78 (s, 3 H), 1.58 (s, 9 H).

tert-Butyl (5-(((3*S*)-4-(4-(methylsulfinyl)phenyl)-3-(1-propyn-1-yl)-1-piperazinyl)sulfonyl)-2-pyridinyl)carbamate (0.10 g, 0.19 mmol) was deprotected according to the general Boc-deprotection method to afford 5-(((3*S*)-4-(4-(methylsulfinyl)phenyl)-3-(1-propyn-1-yl)-1-piperazinyl)sulfonyl)-2-pyridinamine (**14**) (0.040 g, 50%) as a pale-brown solid. MS (ESI positive ion) *m/z*: calcd for $C_{19}H_{22}N_4O_3S_2$, 418.113; found, 419.0 (M + 1). ^{1}H NMR (400 MHz, DMSO-d_6) δ 8.24 (d, J = 2.5 Hz, 1 H), 7.64 (dd, J = 2.5, 8.8 Hz, 1 H), 7.54 (d, J = 8.8 Hz, 2 H), 7.11 (d, J = 8.8 Hz, 2 H), 7.05 (s, 2 H), 6.54 (d, J = 8.8 Hz, 1 H), 4.87 (br s, 1 H), 3.68−3.55 (m, 3 H), 3.15−3.06 (m, 1 H), 2.68 (s, 3 H), 2.43−2.30 (m, 2 H), 1.75 (d, J = 1.4 Hz, 3 H).

4-((2*S*)-4-((6-Amino-3-pyridinyl)sulfonyl)-2-(1-propyn-1-yl)-1-piperazinyl)-*N*-methylbenzenesulfonamide (15). Benzyl (3*S*)-3-(1-propyn-1-yl)-1-piperazinecarboxylate (**4**) (1.00 g, 3.87 mmol) and 4-bromo-*N*-methylbenzenesulfonamide[13] (1.45 g, 5.81 mmol) were coupled according to the general amination method using RuPhos (0.090 g, 0.194 mmol), RuPhos first generation precatalyst (0.158 g, 0.194 mmol), and 1,4-dioxane to afford benzyl (3*S*)-4-(4-(methylsulfamoyl)phenyl)-3-(1-propyn-1-yl)-1-piperazinecarboxylate (0.235 g, 14%) as an amber oil. MS (ESI positive ion) *m/z*: calcd for $C_{22}H_{25}N_3O_4S$, 427.157; found, 428.0 (M + 1). ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.75 (d, J = 8.8 Hz, 2 H), 7.29−7.43 (m, 5 H), 6.99 (d, J = 8.8 Hz, 2 H), 5.09−5.30 (m, 2 H), 4.38−4.52 (m, 1 H), 4.21 (br s, 1 H), 4.25−4.38 (m, 2 H), 3.24−3.53 (m, 3 H), 3.04−3.20 (m, 1 H), 2.65 (d, J = 5.38 Hz, 3 H), 1.69 (br s, 3 H).

Benzyl (3*S*)-4-(4-(methylsulfamoyl)phenyl)-3-(1-propyn-1-yl)-1-piperazinecarboxylate (0.235 g, 0.550 mmol) was deprotected according to the general Cbz-deprotection method A and then treated with *tert*-butyl (5-(chlorosulfonyl)-2-pyridinyl)carbamate (**9**) (0.241 g, 0.825 mmol) according to the general sulfonamide formation method using DIPEA (1.15 mL, 6.60 mmol) to afford *tert*-butyl (5-(((3*S*)-4-(4-(methylsulfamoyl)phenyl)-3-(1-propyn-1-yl)-1-piperazinyl)-sulfonyl)-2-pyridinyl)carbamate (0.160 g, 53% over two steps), as a white solid. MS (ESI positive ion) *m/z*: calcd for $C_{24}H_{31}N_5O_6S_2$, 549.172; found, 550.1 (M + 1).

tert-Butyl (5-(((3*S*)-4-(4-(methylsulfamoyl)phenyl)-3-(1-propyn-1-yl)-1-piperazinyl)sulfonyl)-2-pyridinyl)carbamate (0.160 g, 0.291 mmol) was deprotected according to the general Boc-deprotection method to afford 4-((2*S*)-4-((6-amino-3-pyridinyl)sulfonyl)-2-(1-propyn-1-yl)-1-piperazinyl)-*N*-methylbenzenesulfonamide (**15**) (0.062 g, 47%) as an off-white solid. MS (ESI positive ion) *m/z*: calcd for $C_{19}H_{23}N_5O_6S_2$, 449.119; found, 450.0 (M + 1). ^{1}H NMR (400 MHz, DMSO-d_6) δ 8.27 (d, J = 2.4 Hz, 1 H), 7.67 (dd, J = 8.8, 2.54 Hz, 1 H), 7.62 (d, J = 9.0 Hz, 2 H), 7.17 (q, J = 5.19 Hz, 1 H), 7.10 (d, J = 9.0 Hz, 4 H), 6.57 (d, J = 9.0 Hz, 1 H), 4.93 (br s, 1 H),

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3.72 (d, J = 12.1 Hz, 1 H), 3.59−3.69 (m, 2 H), 3.09−3.19 (m, 1 H), 2.58 (dd, J = 11.5, 3.33 Hz, 1 H), 2.32−2.46 (m, 4 H), 1.77 (d, J = 2.0 Hz, 3 H).

4-((2*S*)-4-((6-Amino-3-pyridinyl)sulfonyl)-2-(1-propyn-1-yl)-1-piperazinyl)-*N*-cyclopropylbenzenesulfonamide (16). (3*S*)-1-Benzyl-3-(1-propyn-1-yl)piperazine (**5**) (0.460 g, 2.18 mmol) and 4-bromo-*N*-cyclopropylbenzenesulfonamide[13] (0.500 g, 1.81 mmol) were coupled according to the general amination method using RuPhos (0.042 g, 0.090 mmol), RuPhos first generation precatalyst (0.042 g, 0.055 mmol), and 1,4-dioxane to give 4-((2*S*)-4-benzyl-2-(1-propyn-1-yl)-1-piperazinyl)-*N*-cyclopropylbenzenesulfonamide (0.350 g, 47%) as a white solid. MS (ESI positive ion) m/z: calcd for $C_{23}H_{27}N_3O_2S$, 409.182; found, 410.1 (M + 1). ^{1}H NMR (300 MHz, DMSO-d_6) δ 7.66−7.57 (m, 3 H), 7.36 (d, J = 6.6 Hz, 4 H), 7.30−7.23 (m, 1 H), 7.08 (d, J = 8.8 Hz, 2 H), 4.78 (bs, 1 H), 3.63 (d, J = 13.6 Hz, 2 H), 3.51 (d, J = 13.6 Hz, 1 H), 3.09 (dd, J = 13.0, 10.0 Hz, 1 H), 2.92 (d, J = 11.0 Hz, 2 H), 2.33−2.12 (m, 2 H), 2.04 (dtt, J = 10.9, 7.5, 3.8 Hz, 1 H), 1.74 (d, J = 2.1 Hz, 3 H), 0.54−0.29 (m, 4 H).

4-((2*S*)-4-Benzyl-2-(1-propyn-1-yl)-1-piperazinyl)-*N*-cyclopropylbenzenesulfonamide (0.350 g, 0.855 mmol) was deprotected according to the general Bn-deprotection method to afford *N*-cyclopropyl-4-((2*S*)-2-(1-propyn-1-yl)-1-piperazinyl)benzenesulfonamide (0.25 g, crude) as a pale-brown residue that was used in the next step without purification. MS (ESI positive ion) m/z: calcd for $C_{16}H_{21}N_3O_2S$, 319.135; found, 319.8 (M + 1).

N-Cyclopropyl-4-((2*S*)-2-(1-propyn-1-yl)-1-piperazinyl)-benzenesulfonamide (0.250 g, 0.783 mmol) was treated with *tert*-butyl (5-(chlorosulfonyl)-2-pyridinyl)carbamate (**9**) (0.340 g, 1.17 mmol) according to the general sulfonamide formation method using pyridine (1.2 mL, 14.8 mmol) to give *tert*-butyl (5-(((3*S*)-4-(4-(cyclopropylsulfamoyl)phenyl)-3-(1-propyn-1-yl)-1-piperazinyl)-sulfonyl)-2-pyridinyl)carbamate (0.200 g, 41% over two steps) as a white solid. MS (ESI positive ion) m/z: calcd for $C_{26}H_{33}N_5O_6S_2$, 575.187; found, 576 (M + 1). ^{1}H NMR (400 MHz, DMSO-d_6) δ 10.53 (s, 1 H), 8.64−8.56 (m, 1 H), 8.11 (dd, J = 9.0, 2.5 Hz, 1 H), 8.04 (dd, J = 9.0, 0.8 Hz, 1 H), 7.71−7.58 (m, 3 H), 7.17−7.03 (m, 2 H), 4.99−4.87 (m, 1 H), 3.79−3.63 (m, 3 H), 3.13 (td, J = 12.2, 3.2 Hz, 1 H), 2.64 (dd, J = 11.5, 3.3 Hz, 1 H), 2.03−1.95 (m, 1 H), 1.75 (d, J = 2.1 Hz, 3 H), 1.48 (s, 9 H), 0.43 (dt, J = 7.9, 3.3 Hz, 2 H), 0.36−0.30 (m, 2 H). One proton was obscured by solvent peak.

tert-Butyl (5-(((3*S*)-4-(4-(cyclopropylsulfamoyl)phenyl)-3-(1-propyn-1-yl)-1-piperazinyl)sulfonyl)-2-pyridinyl)carbamate (0.200 g, 0.347 mmol) was deprotected according to the general Boc-deprotection method to give 4-((2*S*)-4-((6-amino-3-pyridinyl)-sulfonyl)-2-(1-propyn-1-yl)-1-piperazinyl)-*N*-cyclopropyl-benzenesulfonamide (**16**) (0.100 g, 62%) as a white solid. MS (ESI positive ion) m/z: calcd for $C_{21}H_{25}N_5O_4S_2$, 475.135; found, 476.1 (M + 1). ^{1}H NMR (400 MHz, DMSO-d_6) δ 8.24 (d, J = 2.5 Hz, 1 H), 7.68−7.58 (m, 4 H), 7.12−7.02 (m, 4 H), 6.53 (d, J = 8.9 Hz, 1 H), 4.99−4.87 (m, 1 H), 3.76−3.53 (m, 3 H), 3.19−3.06 (m, 1 H), 2.37 (td, J = 11.9, 11.5, 3.0 Hz, 2 H), 2.02 (dq, J = 6.8, 3.4 Hz, 1 H), 1.75 (d, J = 2.1 Hz, 3 H), 0.43 (dt, J = 6.4, 3.1 Hz, 2 H), 0.37−0.28 (m, 2 H).

4-((2*S*)-4-((6-Amino-3-pyridinyl)sulfonyl)-2-(1-propyn-1-yl)-1-piperazinyl)-*N*-(cyclopropylmethyl)benzenesulfonamide (17). (3*S*)-1-Benzyl-3-(1-propyn-1-yl)piperazine (**5**) (0.440 g, 2.08 mmol) and 4-bromo-*N*-(cyclopropylmethyl)benzenesulfonamide[13] (0.500 g, 1.73 mmol) were coupled according to the general amination method using RuPhos (0.040 g, 0.086 mmol), RuPhos first generation precatalyst (0.040 g, 0.051 mmol), and 1,4-dioxane to give 4-((2*S*)-4-benzyl-2-(1-propyn-1-yl)-1-piperazinyl)-*N*-(cyclopropylmethyl)benzenesulfonamide (0.300 g, 41%) as a white solid. MS (ESI positive ion) m/z: calcd for $C_{24}H_{29}N_3O_2S$, 423.198; found, 424.2 (M + 1). ^{1}H NMR (300 MHz, $CDCl_3$) δ 7.78−7.69 (m, 2 H), 7.46−7.25 (m, 4 H), 7.02−6.91 (m, 3 H), 4.53−4.35 (m, 2 H), 3.74 (d, J = 13.5 Hz, 1 H), 3.62−3.44 (m, 2 H), 3.35 (td, J = 11.7, 3.1 Hz, 1 H), 3.08−2.92 (m, 2 H), 2.87−2.72 (m, 2 H), 2.44−2.27 (m, 2 H), 1.80 (d, J = 2.2 Hz, 3 H), 1.28 (d, J = 8.4 Hz, 1 H), 0.89 (qd, J = 8.0, 3.2 Hz, 2 H), 0.53−0.40 (m, 2 H).

(4-((2*S*)-4-Benzyl-2-(1-propyn-1-yl)-1-piperazinyl)-*N*-(cyclopropylmethyl)benzenesulfonamide (0.200 g, 0.47 mmol) was deprotected according to the general Bn-deprotection method to obtain *N*-(cyclopropylmethyl)-4-((2*S*)-2-(1-propyn-1-yl)-1-piperazinyl)benzenesulfonamide (0.15 g, crude) as a pale-brown residue. This material was used in the next step without purification. MS (ESI positive ion) m/z: calcd for $C_{17}H_{23}N_3O_2S$, 333.151; found, 333.9 (M + 1).

N-(Cyclopropylmethyl)-4-((2*S*)-2-(1-propyn-1-yl)-1-piperazinyl)-benzenesulfonamide (0.150 g, 0.45 mmol) was treated with *tert*-butyl (5-(chlorosulfonyl)-2-pyridinyl)carbamate (**9**) (0.197 g, 0.67 mmol) according to the general sulfonamide formation method using pyridine (0.75 mL, 9.3 mmol) to give *tert*-butyl (5-(((3*S*)-4-(4-((cyclopropylmethyl)sulfamoyl)phenyl)-3-(1-propyn-1-yl)-1-piperazinyl)sulfonyl)-2-pyridinyl)carbamate (0.110 g, 41%) as a white solid. MS (ESI positive ion) m/z: calcd for $C_{27}H_{35}N_5O_6S_2$, 589.203; found, 490.1 (M − Boc + 1). ^{1}H NMR (400 MHz, DMSO-d_6) δ 10.54 (s, 1 H), 8.62 (dd, J = 2.5, 0.8 Hz, 1 H), 8.13 (dd, J = 8.9, 2.5 Hz, 1 H), 8.06 (dd, J = 9.0, 0.8 Hz, 1 H), 7.68−7.59 (m, 2 H), 7.50 (t, J = 6.0 Hz, 1 H), 7.14−7.05 (m, 2 H), 4.95 (s, 1 H), 3.83−3.65 (m, 3 H), 3.13 (td, J = 12.3, 3.3 Hz, 1 H), 2.75−2.57 (m, 3 H), 1.76 (d, J = 2.1 Hz, 3 H), 1.50 (s, 9 H), 0.92−0.73 (m, 2 H), 0.41−0.29 (m, 2 H), 0.09−0.03 (m, 2 H).

tert-Butyl (5-(((3*S*)-4-(4-((cyclopropylmethyl)sulfamoyl)phenyl)-3-(1-propyn-1-yl)-1-piperazinyl)sulfonyl)-2-pyridinyl)carbamate (0.110 g, 0.186 mmol) was deprotected according to the general Boc-deprotection method to give 4-((2*S*)-4-benzyl-2-(1-propyn-1-yl)-1-piperazinyl)-*N*-(cyclopropylmethyl)benzenesulfonamide (**17**) (0.050 g, 55%) as a white solid. MS (ESI positive ion) m/z: calcd for $C_{22}H_{27}N_5O_4S_2$, 489.150; found, 489.9 (M+1). ^{1}H NMR (400 MHz, DMSO-d_6) δ 8.26 (d, J = 2.6 Hz, 1 H), 7.69−7.60 (m, 3 H), 7.48 (s, 1 H), 7.12−7.02 (m, 4 H), 6.61−6.52 (m, 1 H), 4.94 (s, 1 H), 3.77−3.56 (m, 3 H), 3.13 (td, J = 12.1, 3.0 Hz, 1 H), 2.59 (td, J = 6.7, 3.4 Hz, 2 H), 2.46−2.32 (m, 2 H), 1.77 (d, J = 2.1 Hz, 3 H), 0.84−0.72 (m, 1 H), 0.39−0.28 (m, 2 H), 0.08−0.03 (m, 2 H).

4-((2*S*)-4-((6-Amino-3-pyridinyl)sulfonyl)-2-(1-propyn-1-yl)-1-piperazinyl)-*N*-phenylbenzenesulfonamide (18). Benzyl (3*S*)-3-(1-propyn-1-yl)-1-piperazinecarboxylate (**4**) (0.290 g, 1.12 mmol) and 4-bromo-*N*-phenylbenzenesulfonamide[13] (0.350 g, 1.12 mmol) were coupled according to the general amination method using JohnPhos (0.016 g, 0.056 mmol), $Pd_2(dba)_3$ (0.030 g, 0.033 mmol), and toluene (5 mL) to give benzyl (3*S*)-4-(4-(phenylsulfamoyl)-phenyl)-3-(1-propyn-1-yl)-1-piperazinecarboxylate (0.170 g, 31%) as a white solid. MS (ESI positive ion) m/z: calcd for $C_{27}H_{27}N_3O_4S$, 489.172; found, 490.0 (M + 1). ^{1}H NMR (300 MHz, DMSO-d_6) δ 10.07 (s, 1 H), 7.63−7.54 (m, 2 H), 7.40−7.35 (m, 3 H), 7.29−7.17 (m, 4 H), 7.09−6.80 (m, 5 H), 5.2 5−5.05 (m, 2 H), 4.78 (s, 1 H), 4.12−3.95 (m, 2 H), 3.60 (s, 1 H), 2.98−3.1 (m, 2 H), 1.63 (d, J = 1.9 Hz, 3 H). One proton was obscured under solvent peak.

Benzyl (3*S*)-4-(4-(phenylsulfamoyl)phenyl)-3-(1-propyn-1-yl)-1-piperazinecarboxylate (0.170 g, 0.34 mmol) was deprotected according to the general Cbz-deprotection method A to give *N*-phenyl-4-((2*S*)-2-(1-propyn-1-yl)-1-piperazinyl)benzenesulfonamide, the title compound (0.100 g, crude), as a pale-brown solid. The material was used in the next step without purification. MS (ESI positive ion) m/z: calcd for $C_{19}H_{21}N_3O_2S$, 355.135; found, 356.1 (M + 1).

N-Phenyl-4-((2*S*)-2-(1-propyn-1-yl)-1-piperazinyl)-benzenesulfonamide (0.100 g, 0.28 mmol) was treated with *tert*-butyl (5-(chlorosulfonyl)-2-pyridinyl)carbamate (**9**) (0.120 g, 0.42 mmol) according to the general sulfonamide formation method using pyridine (0.5 mL, 6.2 mmol) to give *tert*-butyl (5-(((3*S*)-4-(4-(phenylsulfamoyl)phenyl)-3-(1-propyn-1-yl)-1-piperazinyl)sulfonyl)-2-pyridinyl)carbamate (0.060 g, 28% over two steps) as a white solid. MS (ESI positive ion) m/z: calcd for $C_{29}H_{33}N_5O_6S_2$, 611.187; found, 612.3 (M + 1). ^{1}H NMR (300 MHz, $CDCl_3$) δ 8.65 (d, J = 2.3 Hz, 1 H), 8.14 (d, J = 8.8 Hz, 1 H), 8.03 (dd, J = 8.9, 2.4 Hz, 1 H), 7.75−7.58 (m, 2 H), 7.26 (m, 3 H), 7.13−7.04 (m, 2 H), 6.87 (d, J = 8.8 Hz, 2 H), 6.47 (s, 1 H), 4.49 (s, 1 H), 3.90−3.78 (m, 2 H), 3.50 (br d, J = 12.4 Hz, 1 H), 3.46−3.31 (m, 1 H), 2.79 (dd, J = 11.1, 3.0 Hz, 1 H),

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2.72−2.58 (m, 1 H), 1.78 (d, J = 2.1 Hz, 3 H), 1.58 (s, 9 H). One exchangeable proton was not observed.

tert-Butyl (5-(((3*S*)-4-(4-(phenylsulfamoyl)phenyl)-3-(1-propyn-1-yl)-1-piperazinyl)sulfonyl)-2-pyridinyl)carbamate (0.060 g, 0.098 mmol) was deprotected according to the general Boc-deprotection method to give 4-((2*S*)-4-((6-amino-3-pyridinyl)sulfonyl)-2-(1-propyn-1-yl)-1-piperazinyl)-*N*-phenylbenzenesulfonamide (**18**) (0.013 g, 18%) as a white solid. MS (ESI positive ion) m/z: calcd for $C_{24}H_{25}N_5O_4S_2$, 511.135; found, 512.1 (M + 1). ^{1}H NMR (400 MHz, CD_3OD) δ 8.18 (d, J = 2.6 Hz, 1 H), 7.62 (dd, J = 8.9, 2.5 Hz, 1 H), 7.52−7.44 (m, 2 H), 7.09−7.05 (m, 2 H), 7.00−6.82 (m, 5 H), 6.51 (d, J = 9.0 Hz, 1 H), 4.61 (s, 1 H), 3.62 (br d, J = 11.2 Hz, 2 H), 3.49 (br d, J = 12.4 Hz, 1 H), 3.12 (td, J = 12.4, 3.2 Hz, 1 H), 2.58 (dd, J = 11.6, 3.2 Hz, 1 H), 2.42 (td, J = 11.6, 3.2 Hz, 1 H), 1.63 (d, J = 2.1 Hz, 3 H).

5-(((3*S*)-4-(4-(*N*-Methyl-*S*-(trifluoromethyl)sulfonimidoyl)-phenyl)-3-(1-propyn-1-yl)-1-piperazinyl)sulfonyl)-2-pyridinamine (19). (3*S*)-1-Benzyl-3-(1-propyn-1-yl)piperazine (**5**) (0.250 g, 1.15 mmol) and 1-bromo-4-(*N*-methyl-*S*-(trifluoromethyl)-sulfonimidoyl)benzene[13] (0.350 g, 1.15 mmol) were coupled according to the general amination method using RuPhos (0.016 g, 0.034 mmol), $Pd_2(dba)_3$ (0.055 g, 0.058 mmol), and 1,4-dioxane to give (2*S*)-4-benzyl-1-(4-(*N*-methyl-*S*-(trifluoromethyl)sulfonimidoyl)-phenyl)-2-(1-propyn-1-yl)piperazine (0.300 g, 59%) as a white solid. MS (ESI positive ion) m/z: calcd for $C_{22}H_{24}F_3N_3OS$, 435.159; found, 436.0 (M + 1). ^{1}H NMR (300 MHz, $CDCl_3$) δ 7.88 (d, J = 8.8 Hz, 2 H), 7.40 (d, J = 7.2 Hz, 2 H), 7.34 (dd, J = 7.2 Hz, 2 H), 7.28−7.26 (m, 1 H), 6.98 (d, J = 9.2 Hz, 2 H), 4.52 (s, 1 H), 3.72 (d, J = 13.2 Hz, 1 H), 3.60−3.53 (m, 2 H), 3.42−3.35 (m, 1 H), 3.06 (s, 3 H), 3.05−2.98 (m, 2 H), 2.36−2.39 (m, 2 H), 1.81 (d, J = 1.5 Hz, 3 H).

(2*S*)-4-Benzyl-1-(4-(*N*-methyl-*S*-(trifluoromethyl)sulfonimidoyl)-phenyl)-2-(1-propyn-1-yl)piperazine (0.300 g, 0.690 mmol) was deprotected according to the general Bn-deprotection method to afford (2*S*)-1-(4-(*N*-methyl-*S*-(trifluoromethyl)sulfonimidoyl)phenyl)-2-(1-propyn-1-yl)piperazine (0.2 g, crude), which was used in the next step without purification. MS (ESI positive ion) m/z: calcd for $C_{15}H_{18}F_3N_3OS$, 345.112; found, 345.9 (M + 1).

(2*S*)-1-(4-(*N*-Methyl-*S*-(trifluoromethyl)sulfonimidoyl)phenyl)-2-(1-propyn-1-yl)piperazine (0.220 g, 0.636 mmol) was treated with *tert*-butyl (5-(chlorosulfonyl)-2-pyridinyl)carbamate (**9**) (0.190 g, 0.636 mmol) according to the general sulfonamide formation method using Et_3N (0.2 mL, 2.5 mmol) in THF (15 mL) instead of DCM to afford *tert*-butyl (5-(((3*S*)-4-(4-(*N*-methyl-*S*-(trifluoromethyl)-sulfonimidoyl)phenyl)-3-(1-propyn-1-yl)-1-piperazinyl)sulfonyl)-2-pyridinyl)carbamate (0.250 g, 60% over two steps) as a white solid. MS (ESI positive ion) m/z: calcd for $C_{25}H_{30}F_3N_5O_5S_2$, 601.164; found, 602.2 (M + 1). ^{1}H NMR (300 MHz, DMSO-d_6) δ 10.31 (s, 1 H), 8.59 (d, J = 1.8 Hz, 1 H), 8.13−8.08 (m, 1 H), 8.04−8.01 (d, J = 9.0 Hz, 1 H), 7.82 (d, J = 9.0 Hz, 2 H), 7.18 (d, J = 9.0 Hz. 2 H), 5.03 (bs, 1 H), 3.88 (bd, J = 12.3 Hz, 1 H), 3.75−3.67 (m, 2 H), 3.31−3.05 (m, 2 H), 2.93 (s, 3 H), 2.67−2.64 (m, 1 H), 1.76 (d, J = 1.5 Hz, 3 H), 1.47 (s, 9 H).

tert-Butyl (5-(((3*S*)-4-(4-(*N*-methyl-*S*-(trifluoromethyl)-sulfonimidoyl)phenyl)-3-(1-propyn-1-yl)-1-piperazinyl)sulfonyl)-2-pyridinyl)carbamate (0.15 g, 0.25 mmol) was deprotected according to the general Boc-deprotection method to give 5-(((3*S*)-4-(4-(*N*-methyl-*S*-(trifluoromethyl)sulfonimidoyl)phenyl)-3-(1-propyn-1-yl)-1-piperazinyl)sulfonyl)-2-pyridinamine (**19**) (0.045 g, 36%) as a white solid. MS (ESI positive ion) m/z: calcd for $C_{20}H_{22}F_3N_5O_3S_2$, 501.112; found, 502 (M + 1). ^{1}H NMR (400 MHz, $CDCl_3$) δ 8.51 (d, J = 2.4 Hz, 1 H), 7.91 (d, J = 8.8 Hz, 2 H), 7.78 (dd, J = 8.8, 2 Hz, 1 H), 6.99 (d, J = 8.0 Hz, 2 H), 6.55 (d, J = 8.8 Hz, 1 H), 5.03 (s, 2 H), 4.59 (bs, 1 H), 3.88−3.82 (m, 2 H), 3.7−3.63 (m, 1 H), 3.53−3.46 (m, 1 H), 3.07 (s, 3 H), 2.8 (dd, J = 11.2, 3.2 Hz, 1 H), 2.71−2.63 (m, 1 H), 1.81 (d, J = 2 Hz, 3 H).

5-(((3*S*)-4-(4-(Methylsulfonyl)phenyl)-3-(1-propyn-1-yl)-1-piperazinyl)sulfonyl)-2-pyridinamine (20). Benzyl 3-(1-propyn-1-yl)-1-piperazinecarboxylate (**6**) (0.450 g, 1.74 mmol) and 1-bromo-4-(methylsulfonyl)benzene (0.500 g, 2.12 mmol) were coupled according to the general amination method using BINAP (0.032 g, 0.05 mmol), $Pd(OAc)_2$ (0.011 g, 0.04 mmol), and toluene to obtain benzyl 4-(4-(methylsulfonyl)phenyl)-3-(1-propyn-1-yl)-1-piperazine-carboxylate (0.220 g, 25%) as a white solid. MS (ESI positive ion) m/z: calcd for $C_{22}H_{24}N_2O_4S$, 412.146; found, 413.1 (M + 1).

Benzyl 4-(4-(methylsulfonyl)phenyl)-3-(1-propyn-1-yl)-1-piperazinecarboxylate (0.22 g, 0.53 mmol) was deprotected according to the general Cbz-deprotection method A to obtain 1-(4-(methylsulfonyl)-phenyl)-2-(prop-1-yn-1-yl)piperazine (0.13 g, crude) as a pale-brown solid. The material was used in the next step without purification. MS (ESI positive ion) m/z: calcd for $C_{14}H_{18}N_2O_2S$, 278.109; found, 279.1 (M + 1).

1-(4-(Methylsulfonyl)phenyl)-2-(1-propyn-1-yl)piperazine (0.12 g, 0.43 mmol) was treated with *tert*-butyl (5-(chlorosulfonyl)-2-pyridinyl)carbamate (**9**) (0.127 g, 0.43 mmol) according to the general sulfonamide formation method using Et_3N (0.2 mL, 1.9 mmol) to afford *tert*-butyl (5-((4-(4-(methylsulfonyl)phenyl)-3-(1-propyn-1-yl)-1-piperazinyl)sulfonyl)-2-pyridinyl)carbamate (0.080 g, 35%) as a white solid. MS (ESI positive ion) m/z: calcd for $C_{24}H_{30}N_4O_6S_2$, 534.161; found, 534.5 (M + 1). ^{1}H NMR (400 MHz, DMSO-d_6) δ 8.59 (bs,1 H), 8.09−8.01 (m, 2 H), 7.72 (d, J = 6.6, 2.4 Hz, 2 H), 7.12 (d, J = 6.9 Hz, 2 H), 7.03 (s, 2 H), 4.96 (s, 1 H), 3.76−3.67 (m, 3 H), 3.11 (m, 4 H), 2.62 (d, J = 7.8 Hz, 1 H), 1.74 (s, 3 H), 1.47 (s, 9 H).

tert-Butyl (5-((4-(4-(methylsulfonyl)phenyl)-3-(1-propyn-1-yl)-1-piperazinyl)sulfonyl)-2-pyridinyl)carbamate (0.080 g, 0.15 mmol) was deprotected according to the general Boc-deprotection method to afford 5-4-(4-(methylsulfonyl)phenyl)-3-(1-propyn-1-yl)-1-piperazinyl)sulfonyl)-2-pyridinamine (0.030 g, 46%) as a white solid. MS (ESI positive ion) m/z: calcd for $C_{19}H_{22}N_3O_4S_2$, 434.108; found, 435.3 (M + 1). ^{1}H NMR (400 MHz, DMSO-d_6) δ 8.24 (d, J = 2.5 Hz, 1 H), 7.72 (d, J = 8.8 Hz, 2 H), 7.64 (dd, J = 2.5, 8.8 Hz, 1 H), 7.12 (d, J = 9.1 Hz, 2 H), 7.05 (s, 2 H), 6.53 (d, J = 8.8 Hz, 1 H), 4.97 (br s, 1 H), 3.76 (d, J = 12.2 Hz, 1 H), 3.63 (t, J = 10.1 Hz, 2 H), 3.17 (d, J = 5.3 Hz, 1 H), 3.12 (s, 3 H), 2.42−2.30 (m, 2 H), 1.76 (d, J = 1.4 Hz, 3 H).

The racemic 5-4-(4-(methylsulfonyl)phenyl)-3-(1-propyn-1-yl)-1-piperazinyl)sulfonyl)-2-pyridinamine (0.030 g, 0.069 mmol) was separated using chiral SFC as follows: Sepax OJH (5 μm, 21 mm × 250 mm) column, using 70% carbon dioxide−30% MeOH containing 20 mM ammonia; flow rate was 80 mL/min. This procedure produced 5-(((3*S*)-4-(4-(methylsulfonyl)phenyl)-3-(1-propyn-1-yl)-1-piperazinyl)sulfonyl)-2-pyridinamine (**20**). The absolute stereochemistry at the piperazine carbon next to the N−Ar was assigned to be (*S*) configuration based on the result obtained in the biochemical assay. MS (ESI positive ion) m/z: calcd for $C_{19}H_{22}N_3O_4S_2$, 434.108; found, 434.9 (M + 1). ^{1}H NMR (300 MHz, $CDCl_3$) δ 8.49 (d, J = 1.9 Hz, 1 H), 7.86−7.76 (m, 3 H), 7.04−6.95 (m, 2 H), 6.59 (d, J = 8.8 Hz, 1 H), 5.32 (br s, 2 H), 4.55 (br s, 1 H), 3.90−3.76 (m, 2 H), 3.59 (br s, 1 H), 3.50−3.37 (m, J = 2.9, 11.7 Hz, 1 H), 3.03 (s, 3 H), 2.85 (dd, J = 3.4, 11.5 Hz, 1 H), 2.77−2.65 (m, 1 H), 1.80 (s, 3 H).

4-((2*S*)-4-((6-Amino-3-pyridinyl)sulfonyl)-2-(1-propyn-1-yl)-1-piperazinyl)-*N*-(1-methylethyl)benzenesulfonamide (21). Benzyl 3-(1-propyn-1-yl)-1-piperazinecarboxylate (**6**) (0.370 g, 1.44 mmol) and 4-bromo-*N*-(1-methylethyl)benzenesulfonamide[13] (0.400 g, 1.44 mmol) were coupled according to the general amination method using JohnPhos (0.021 g, 0.072 mmol), $Pd_2(dba)_3$ (0.040 g, 0.043 mmol), and toluene to give benzyl 4-(4-((1-methylethyl)-sulfamoyl)phenyl)-3-(1-propyn-1-yl)-1-piperazinecarboxylate (0.300 g, 46%) as a white solid. MS (ESI positive ion) m/z: calcd for $C_{24}H_{29}N_3O_4S$, 455.188; found, 456.1 (M + 1). ^{1}H NMR (300 MHz, $CDCl_3$) δ 7.75 (br d, J = 8.7 Hz, 2 H), 7.42−7.33 (m, 4 H), 6.97 (br d, J = 9 Hz, 3 H), 5.32−5.10 (m, 2 H), 4.55−4.20 (m, 4 H), 3.53−3.19 (m, 4 H), 3.18−3.03 (m, 1 H), 1.65 (d, J = 2.1 Hz, 3 H), 1.08 (d, J = 6.5 Hz, 6H).

Benzyl 4-(4-((1-methylethyl)sulfamoyl)phenyl)-3-(1-propyn-1-yl)-1-piperazinecarboxylate (0.300 g, 0.65 mmol) was deprotected according to the general Cbz-deprotection method A to give *N*-(1-methylethyl)-4-(2-(1-propyn-1-yl)-1-piperazinyl)benzenesulfonamide (0.2 g, crude) as a pale-brown solid. This material was used in the next

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step without purification. MS (ESI positive ion) m/z: calcd for $C_{16}H_{23}N_3O_2S$, 321.151; found, 322.1 (M + 1).

N-(1-Methylethyl)-4-(2-(1-propyn-1-yl)-1-piperazinyl)-benzenesulfonamide (0.200 g, 0.623 mmol) was treated with *tert*-butyl (5-(chlorosulfonyl)-2-pyridinyl)carbamate (**9**) (0.200 g, 0.68 mmol) according to the general sulfonamide formation method using pyridine (1.0 mL, 12 mmol) to give *tert*-butyl (5-((4-(4-((1-methylethyl)-sulfamoyl)phenyl)-3-(1-propyn-1-yl)-1-piperazinyl)sulfonyl)-2-pyridinyl)carbamate (0.150 g, 40% over two steps) as a white solid. MS (ESI positive ion) m/z: calcd for $C_{26}H_{35}N_5O_6S_2$, 577.203; found, 478.1 (M − Boc + 1). ^{1}H NMR (300 MHz, DMSO-d_6) δ 10.51 (s, 1 H), 8.60 (br s, 1 H), 8.16−7.99 (m, 2 H), 7.62 (br d, J = 8.4 Hz, 2 H), 7.32 (d, J = 7.0 Hz, 1 H), 7.07 (d, J = 8.4 Hz, 2 H), 4.93 (s, 1 H), 3.78−3.62 (m, 3 H), 3.20−3.05 (m, 2 H), 2.64 (br d, J = 8.7 Hz, 1 H), 1.73 (d, J = 1.9 Hz, 3 H), 1.48 (s, 9 H), 0.90 (d, J = 6.4 Hz, 6 H). One proton was obscured under solvent peak.

tert-Butyl (5-((4-(4-((1-methylethyl)sulfamoyl)phenyl)-3-(1-propyn-1-yl)-1-piperazinyl)sulfonyl)-2-pyridinyl)carbamate (0.150 g, 0.25 mmol) was deprotected according to the general Boc-deprotection method to give 4-(4-((6-amino-3-pyridinyl)sulfonyl)-2-(1-propyn-1-yl)-1-piperazinyl)-*N*-(1-methylethyl)benzenesulfonamide (0.080 g, 67%) as a white solid. MS (ESI positive ion) m/z: calcd for $C_{21}H_{27}N_5O_4S_2$, 477.150; found, 478.1 (M + 1). ^{1}H NMR (400 MHz, DMSO-d_6) δ 8.24 (d, J = 2.5 Hz, 1 H), 7.68−7.57 (m, 3 H), 7.31 (d, J = 7.1 Hz, 1 H), 7.11−7.02 (m, 4 H), 6.53 (d, J = 8.9 Hz, 1 H), 4.92 (br s, 1 H), 3.73−3.56 (m, 3 H), 3.20−3.05 (m, 2 H), 2.45−2.35 (m, 1 H), 1.74 (d, J = 2.2 Hz, 3 H), 0.91 (d, J = 6.4 Hz, 6 H). One proton was obscured under solvent peak.

The racemic 4-(4-((6-amino-3-pyridinyl)sulfonyl)-2-(1-propyn-1-yl)-1-piperazinyl)-*N*-(1-methylethyl)benzenesulfonamide (0.030 g, 0.063 mmol) was separated using chiral SFC as follows: Chiralpak AS-H (5 μm, 21 mm × 250 mm) column, using 70% carbon dioxide−30% MeOH containing 20 mM ammonia; flow rate was 70 mL/min. This procedure produced 4-((2*S*)-4-((6-amino-3-pyridinyl)sulfonyl)-2-(1-propyn-1-yl)-1-piperazinyl)-*N*-(1-methylethyl)-benzenesulfonamide (**21**). The absolute stereochemistry at the piperazine carbon next to the N−Ar was assigned to be (*S*) configuration based on the result obtained in the biochemical assay. MS (ESI positive ion) m/z: calcd for $C_{21}H_{27}N_5O_4S_2$, 477.150; found, 477.9 (M + 1). ^{1}H NMR (300 MHz, $CDCl_3$) δ 8.50 (d, J = 2.3 Hz, 1 H), 7.79 (dd, J = 2.3, 8.8 Hz, 1 H), 7.75 (d, J = 9.1 Hz, 2 H), 6.95 (d, J = 9.1 Hz, 2 H), 6.55 (d, J = 8.8 Hz, 1 H), 5.12 (s, 2 H), 4.52 (br s, 1 H), 4.14 (d, J = 7.5 Hz, 1 H), 3.88−3.73 (m, 2 H), 3.59−3.35 (m, 3 H), 2.84 (dd, J = 3.4, 11.3 Hz, 1 H), 2.70 (dt, J = 3.5, 11.3 Hz, 1 H), 1.79 (d, J = 2.0 Hz, 3 H), 1.08 (d, J = 6.6 Hz, 6 H).

5-(((3*S*)-3-(1-Propyn-1-yl)-4-(4-(*S*-(trifluoromethyl)-sulfonimidoyl)phenyl)-1-piperazinyl)sulfonyl)-2-pyridinamine (22). Benzyl (3*S*)-3-(1-propyn-1-yl)-1-piperazinecarboxylate (**4**) (0.684 g, 2.65 mmol) and 1-bromo-4-(*S*-(trifluoromethyl)-sulfonimidoyl)benzene[13] (0.758 g, 2.63 mmol) were coupled according to the general amination method using RuPhos (0.123 g, 0.263 mmol), RuPhos first generation precatalyst (0.214 g, 0.262 mmol), and 1,4-dioxane to afford benzyl (3*S*)-3-(1-propyn-1-yl)-4-(4-(*S*-(trifluoromethyl)sulfonimidoyl)phenyl)-1-piperazinecarboxylate (0.891 g, 73%) as a light-yellow foam. MS (ESI positive ion) m/z: calcd for $C_{22}H_{22}F_3N_3O_3S$, 465.133; found, 466.1 (M + 1). ^{1}H NMR (300 MHz, $CDCl_3$) δ 7.98 (d, J = 9.1 Hz, 2 H), 7.45−7.29 (m, 5 H), 7.03 (d, J = 9.2 Hz, 2 H), 5.21 (d, J = 17.0 Hz, 2 H), 4.50 (br s, 1 H), 4.33 (d, J = 11.4 Hz, 2 H), 3.61 (d, J = 11.3 Hz, 1 H), 3.50−3.08 (m, 4 H), 1.72 (br s, 3 H).

Benzyl (3*S*)-3-(1-propyn-1-yl)-4-(4-(*S*-(trifluoromethyl)-sulfonimidoyl)phenyl)-1-piperazinecarboxylate (0.890 g, 1.91 mmol) was deprotected according to the general Cbz-deprotection method A, then treated with *tert*-butyl (5-(chlorosulfonyl)-2-pyridinyl)carbamate (**9**) (0.627 g, 2.14 mmol) according to the general sulfonamide formation method using Et_3N (2.7 mL, 19.4 mmol), and finally deprotected according to the general Boc-deprotection method to afford 5-(((3*S*)-3-(1-propyn-1-yl)-4-(4-(*S*-(trifluoromethyl)-sulfonimidoyl)phenyl)-1-piperazinyl)sulfonyl)-2-pyridinamine (**22**) (0.886 g, 95%) as an off-white foam. MS (ESI positive ion) m/z: calcd for $C_{19}H_{20}F_3N_5O_3S_2$, 487.096; found, 488.1 (M + 1). ^{1}H NMR (300 MHz, $CDCl_3$) δ 8.48 (d, J = 2.2 Hz, 1 H), 7.95 (d, J = 9.1 Hz, 2 H), 7.76 (dd, J = 2.3, 8.8 Hz, 1 H), 6.99 (d, J = 9.1 Hz, 2 H), 6.52 (d, J = 8.8 Hz, 1 H), 5.03 (s, 2 H), 4.58 (br s, 1 H), 3.90−3.75 (m, 2 H), 3.66 (br s, 1 H), 3.54−3.37 (m, 2 H), 2.78 (dd, J = 3.4, 11.5 Hz, 1 H), 2.66 (dt, J = 3.0, 11.6 Hz, 1 H), 1.79 (d, J = 1.8 Hz, 3 H).

5-(((3*S*)-3-(1-Propyn-1-yl)-4-(4-(*R*)-(*S*-(trifluoromethyl)-sulfonimidoyl)phenyl)-1-piperazinyl)sulfonyl)-2-pyridinamine (23) and 5-(((3*S*)-3-(1-Propyn-1-yl)-4-(4-(*S*)-(*S*-(trifluoromethyl)-sulfonimidoyl)phenyl)-1-piperazinyl)sulfonyl)-2-pyridinamine (24). The individual diastereomers were isolated using chiral SFC. The method used was as follows: Chiralpak AS-H column (21 mm × 250 mm, 5 μm) using 30% (240 mM NH_3 in methanol) in supercritical CO_2 (total flow was 70 mL/min). This produced the two diastereomers with diastereomeric and enanteomeric excesses greater than 98%. The absolute stereochemistry around the sulfur in sulfoximine moiety was determined using enantiomeric pure 1-bromo-4-(*S*-(trifluoromethyl)sulfonimidoyl)benzene.[16]

5-(((3*S*)-3-(1-Propyn-1-yl)-4-(4-(*R*)-(*S*-(trifluoromethyl)-sulfonimidoyl)phenyl)-1-piperazinyl)sulfonyl)-2-pyridinamine (**23**). MS (ESI positive ion) m/z: calcd for $C_{19}H_{20}F_3N_5O_3S_2$, 487.096; found, 488.1 (M + 1). ^{1}H NMR (300 MHz, $CDCl_3$) δ 8.48 (d, J = 2.2 Hz, 1 H), 7.95 (d, J = 9.1 Hz, 2 H), 7.77 (d, J = 2.5 Hz, 1 H), 6.99 (d, J = 9.2 Hz, 2 H), 6.52 (d, J = 8.9 Hz, 1 H), 5.00 (s, 2 H), 4.57 (br s, 1 H), 3.91−3.76 (m, 2 H), 3.65 (d, J = 12.7 Hz, 1 H), 3.54−3.38 (m, 2 H), 2.78 (dd, J = 3.6, 11.5 Hz, 1 H), 2.66 (dt, J = 3.3, 11.6 Hz, 1 H), 1.79 (d, J = 2.0 Hz, 3 H).

5-(((3*S*)-3-(1-Propyn-1-yl)-4-(4-(*S*)-(*S*-(trifluoromethyl)-sulfonimidoyl)phenyl)-1-piperazinyl)sulfonyl)-2-pyridinamine (**24**). MS (ESI positive ion) m/z: calcd for $C_{19}H_{20}F_3N_5O_3S_2$, 487.096; found, 488.1 (M + 1). ^{1}H NMR (300 MHz, $CDCl_3$) δ 8.51 (d, J = 2.2 Hz, 1 H), 7.97 (d, J = 9.1 Hz, 2 H), 7.78 (dd, J = 2.5, 8.8 Hz, 1 H), 7.01 (d, J = 9.2 Hz, 2 H), 6.54 (d, J = 8.8 Hz, 1 H), 5.02 (s, 2 H), 4.60 (br s, 1 H), 3.93−3.77 (m, 2 H), 3.72−3.60 (m, 1 H), 3.56−3.40 (m, 2 H), 2.80 (dd, J = 3.6, 11.5 Hz, 1 H), 2.68 (dt, J = 3.4, 11.6 Hz, 1 H), 1.81 (d, J = 2.2 Hz, 3 H).

2-(5-((2*S*)-4-((6-Amino-3-pyridinyl)sulfonyl)-2-(1-propyn-1-yl)-1-piperazinyl)-2-pyridinyl)-1,1,1-trifluoro-2-propanol (25). (3*S*)-1-Benzyl-3-(1-propyn-1-yl)piperazine (**5**) (0.180 g, 0.839 mmol) and 2-(5-bromo-2-pyridinyl)-1,1,1-trifluoro-2-propanol[13] (0.232 g, 0.859 mmol) were coupled according to the general amination method using RuPhos (0.040 g, 0.085 mmol), RuPhos first generation precatalyst (0.072 g, 0.089 mmol), and 1,4-dioxane to afford 2-(5-((2*S*)-4-benzyl-2-(1-propyn-1-yl)-1-piperazinyl)-2-pyridinyl)-1,1,1-trifluoro-2-propanol (0.289 g, 85%) as a light-brown oil. MS (ESI positive ion) m/z: calcd for $C_{22}H_{24}F_3N_3O$, 403.187; found, 404.0 (M + 1). ^{1}H NMR (300 MHz, $CDCl_3$) δ 8.26 (t, J = 2.5 Hz, 1 H), 7.45−7.27 (m, 7 H), 6.23 (s, 1 H), 4.38 (br s, 1 H), 3.73 (d, J = 10.7 Hz, 1 H), 3.58 (s, 1 H), 3.47−3.30 (m, 2 H), 3.06−2.92 (m, 2 H), 2.50−2.32 (m, 2 H), 1.81 (d, J = 2.0 Hz, 3 H), 1.70 (s, 3 H).

2-(5-((2*S*)-4-Benzyl-2-(1-propyn-1-yl)-1-piperazinyl)-2-pyridinyl)-1,1,1-trifluoro-2-propanol (0.289 g, 0.716 mmol) was deprotected according to the general Bn-deprotection method. The crude product was treated with *tert*-butyl (5-(chlorosulfonyl)-2-pyridinyl)carbamate (**9**) (0.238 g, 0.813 mmol) according to the general sulfonamide formation method using Et_3N (1.0 mL, 7.2 mmol) to afford *tert*-butyl (5-(((3*S*)-3-(1-propyn-1-yl)-4-(6-(2,2,2-trifluoro-1-hydroxy-1-methylethyl)-3-pyridinyl)-1-piperazinyl)sulfonyl)-2-pyridinyl)carbamate (0.347 g, 85%) as a white solid. MS (ESI positive ion) m/z: calcd for $C_{25}H_{30}F_3N_5O_5S$, 569.192; found, 570.0 (M + 1). ^{1}H NMR (300 MHz, $CDCl_3$) δ 8.67 (dd, J = 0.7, 2.3 Hz, 1 H), 8.23 (t, J = 2.6 Hz, 1 H), 8.18−8.11 (m, 1 H), 8.07−8.00 (m, 1 H), 7.84 (s, 1 H), 7.42−7.35 (m, 1 H), 7.35−7.28 (m, 1 H), 6.10 (s, 1 H), 4.39 (br s, 1 H), 3.87−3.72 (m, 2 H), 3.48−3.32 (m, 2 H), 2.96−2.86 (m, J = 3.2 Hz, 1 H), 2.82−2.69 (m, 1 H), 1.79 (s, 3 H), 1.68 (s, 3 H), 1.55 (s, 9 H).

tert-Butyl (5-(((3*S*)-3-(1-propyn-1-yl)-4-(6-(2,2,2-trifluoro-1-hydroxy-1-methylethyl)-3-pyridinyl)-1-piperazinyl)sulfonyl)-2-pyridinyl)carbamate (0.347 g, 0.609 mmol) was deprotected according to the general Boc-deprotection method to afford 2-(5-((2*S*)-4-((6-amino-3-pyridinyl)sulfonyl)-2-(1-propyn-1-yl)-1-piperazinyl)-2-pyri-

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dinyl)-1,1,1-trifluoro-2-propanol (**25**) (0.225 g, 79%) as a white solid. MS (ESI positive ion) *m/z*: calcd for $C_{20}H_{22}F_3N_5O_3S$, 469.140; found, 469.9 (M + 1). ^{1}H NMR (300 MHz, $CDCl_3$) δ 8.49 (d, *J* = 2.0 Hz, 1 H), 8.23 (t, *J* = 2.6 Hz, 1 H), 7.77 (dd, *J* = 2.5, 8.8 Hz, 1 H), 7.41−7.28 (m, 2 H), 6.53 (d, *J* = 8.8 Hz, 1 H), 6.10 (s, 1 H), 5.00 (s, 2 H), 4.38 (br s, 1 H), 3.81−3.68 (m, 2 H), 3.44−3.33 (m, 2 H), 2.88 (dd, *J* = 3.4, 11.4 Hz, 1 H), 2.79−2.67 (m, 1 H), 1.78 (d, *J* = 2.2 Hz, 3 H), 1.68 (s, 3 H).

2-(6-((2*S*)-4-((6-Amino-3-pyridinyl)sulfonyl)-2-(1-propyn-1-yl)-1-piperazinyl)-3-pyridinyl)-1,1,1,3,3,3-hexafluoro-2-propanol (26). Benzyl (3*S*)-3-(1-propyn-1-yl)-1-piperazinecarboxylate (**4**) (0.129 g, 0.500 mmol) and a mixture of 2-(6-bromo-3-pyridinyl)-1,1,1,3,3,3-hexafluoro-2-propanol and 2-(6-chloro-3-pyridinyl)-1,1,1,3,3,3-hexafluoro-2-propanol[13] (~1:1, 0.160 g, 0.53 mmol) were coupled according to the general amination method using RuPhos and RuPhos first generation precatalyst premix (1:1) (0.041 g, 0.034 mmol) and 1,4-dioxane to afford benzyl (3*S*)-3-(1-propyn-1-yl)-4-(5-(2,2,2-trifluoro-1-hydroxy-1-(trifluoromethyl)ethyl)-2-pyridinyl)-1-piperazinecarboxylate (0.135 g, 55%) as a pale-yellow foam. MS (ESI positive ion) *m/z*: calcd for $C_{23}H_{21}F_6N_3O_3$: 501.149; found, 502.1 (M + 1). ^{1}H NMR (300 MHz, $CDCl_3$) δ 8.54 (d, *J* = 2.0 Hz, 1 H), 7.80 (dd, *J* = 2.1, 9.3 Hz, 1 H), 7.46−7.30 (m, 5 H), 6.71 (d, *J* = 9.1 Hz, 1 H), 5.22 (d, *J* = 17.8 Hz, 3 H), 4.44−4.18 (m, 2 H), 4.03 (br s, 1 H), 3.48 (s, 1 H), 3.45−3.30 (m, 1 H), 3.29−2.92 (m, 2 H), 1.71 (br s, 3 H).

Benzyl (3*S*)-3-(1-propyn-1-yl)-4-(5-(2,2,2-trifluoro-1-hydroxy-1-(trifluoromethyl)ethyl)-2-pyridinyl)-1-piperazinecarboxylate (0.135 g, 0.269 mmol) was deprotected according to the general Cbz-deprotection method A. The crude product was treated with *tert*-butyl (5-(chlorosulfonyl)-2-pyridinyl)carbamate (**9**) (0.086 g, 0.29 mmol) according to the general sulfonamide formation method using Et_3N (0.40 mL, 2.9 mmol) and then deprotected according to the general Boc-deprotection method to afford 2-(6-((2*S*)-4-((6-amino-3-pyridinyl)sulfonyl)-2-(1-propyn-1-yl)-1-piperazinyl)-3-pyridinyl)-1,1,1,3,3,3-hexafluoro-2-propanol (**26**) (0.125 g, 89%) as a white solid. MS (ESI positive ion) *m/z*: calcd for $C_{20}H_{19}F_6N_5O_3S$, 523.111; found, 524.1 (M + 1). ^{1}H NMR (300 MHz, $CDCl_3$) δ 8.49 (dd, *J* = 2.0, 9.6 Hz, 2 H), 7.79 (dd, *J* = 2.3, 8.8 Hz, 2 H), 6.68 (d, *J* = 9.1 Hz, 1 H), 6.54 (d, *J* = 8.8 Hz, 1 H), 5.25 (br s, 1 H), 5.09 (s, 2 H), 4.21−4.10 (m, 1 H), 3.92−3.77 (m, 2 H), 3.71−3.36 (m, 2 H), 2.71 (dd, *J* = 3.7, 11.3 Hz, 1 H), 2.58 (dt, *J* = 3.2, 11.8 Hz, 1 H), 1.81 (d, *J* = 2.0 Hz, 3 H).

6-((2*S*)-4-((6-Amino-3-pyridinyl)sulfonyl)-2-(1-propyn-1-yl)-1-piperazinyl)-*N*-methyl-3-pyridinesulfonamide (27). Benzyl (3*S*)-3-(1-propyn-1-yl)-1-piperazinecarboxylate (**4**) (0.507 g, 1.96 mmol) and 6-chloro-*N*-methyl-3-pyridinesulfonamide[13] (0.501 g, 2.42 mmol) were coupled according to the general amination procedure using RuPhos and RuPhos first generation precatalyst premix (1:1) (0.235 g, 0.196 mmol) and 1,4-dioxane to afford benzyl (3*S*)-4-(5-(methylsulfamoyl)-2-pyridinyl)-3-(1-propyn-1-yl)-1-piperazinecarboxylate (0.309 g, 37%) as a white solid. MS (ESI positive ion) *m/z*: calcd for $C_{21}H_{24}N_4O_4S$, 428.152; found, 429.2 (M + 1). ^{1}H NMR (300 MHz, DMSO-d_6) δ 8.44 (d, *J* = 2.3 Hz, 1 H), 8.23 (d, *J* = 2.3 Hz, 1 H), 7.86 (dd, *J* = 2.5, 9.1 Hz, 1 H), 7.63 (dd, *J* = 2.5, 8.9 Hz, 1 H), 7.30 (d, *J* = 3.4 Hz, 1 H), 6.99 (t, *J* = 4.4 Hz, 3 H), 6.52 (d, *J* = 8.9 Hz, 1 H), 5.48 (br s, 1 H), 4.29 (d, *J* = 12.3 Hz, 1 H), 3.65 (d, *J* = 11.1 Hz, 2 H), 3.27−3.13 (m, 2 H), 2.44−2.24 (m, 4 H), 1.78 (d, *J* = 2.0 Hz, 3 H). One exchangeable proton was not observed, and another proton was obscured by the solvent peak.

Benzyl (3*S*)-4-(5-(methylsulfamoyl)-2-pyridinyl)-3-(1-propyn-1-yl)-1-piperazinecarboxylate (0.157 g, 0.366 mmol) was deprotected according to the general Cbz-deprotection method A. The crude product was treated with *tert*-butyl (5-(chlorosulfonyl)-2-pyridinyl)-carbamate (**9**) (0.0865 g, 0.295 mmol) according to the general sulfonamide formation using Et_3N (0.50 mL, 3.6 mmol) and then deprotected according to the general Boc-deprotection method to afford 6-((2*S*)-4-((6-amino-3-pyridinyl)sulfonyl)-2-(1-propyn-1-yl)-1-piperazinyl)-*N*-methyl-3-pyridinesulfonamide (**27**) (0.0247 g, 15% over three steps) as an off-white solid. MS (ESI positive ion) *m/z*: calcd for $C_{18}H_{22}N_6O_4S_2$, 450.114; found, 451.1 (M + 1). ^{1}H NMR (300 MHz, DMSO-d_6) δ 8.44 (d, *J* = 2.3 Hz, 1 H), 8.23 (d, *J* = 2.3 Hz, 1 H), 7.86 (dd, *J* = 2.5, 9.1 Hz, 1 H), 7.63 (dd, *J* = 2.5, 8.9 Hz, 1 H), 7.30 (d, *J* = 3.4 Hz, 1 H), 6.99 (t, *J* = 4.4 Hz, 3 H), 6.52 (d, *J* = 8.9 Hz, 1 H), 5.48 (br s, 1 H), 4.29 (d, *J* = 12.3 Hz, 1 H), 3.65 (d, *J* = 11.1 Hz, 2 H), 3.27−3.13 (m, 2 H), 2.44−2.24 (m, 4 H), 1.78 (d, *J* = 2.0 Hz, 3 H).

2-(6-((2*S*)-4-((6-Amino-3-pyridinyl)sulfonyl)-2-(1-propyn-1-yl)-1-piperazinyl)-3-pyridinyl)-1,1,1-trifluoro-2-propanol (28). Benzyl (3*S*)-3-(1-propyn-1-yl)-1-piperazinecarboxylate (**4**) (1.04 g, 4.03 mmol) and 2-(6-bromo-3-pyridinyl)-1,1,1-trifluoro-2-propanol[13] (1.39 g, 5.15 mmol) were coupled according to the general amination method using RuPhos and RuPhos first generation precatalyst premix (1:1) (0.245 g, 0.205 mmol) and 1,4-dioxane to afford benzyl (3*S*)-3-(1-propyn-1-yl)-4-(5-(2,2,2-trifluoro-1-hydroxy-1-methylethyl)-2-pyridinyl)-1-piperazinecarboxylate (1.63 g, 90%) as a light-yellow foam. MS (ESI positive ion) *m/z*: calcd for $C_{23}H_{24}F_3N_3O_3$, 447.177; found, 448.2 (M + 1). ^{1}H NMR (300 MHz, $CDCl_3$) δ 8.41 (s, 1 H), 7.74 (d, *J* = 9.1 Hz, 1 H), 7.47−7.29 (m, 5 H), 6.68 (d, *J* = 8.9 Hz, 1 H), 5.37−5.05 (m, 3 H), 4.33 (d, *J* = 10.8 Hz, 2 H), 3.97 (d, *J* = 9.9 Hz, 1 H), 3.46−2.92 (m, 3 H), 2.42 (s, 1 H), 1.77 (s, 3 H), 1.70 (br s, 3 H).

Benzyl (3*S*)-3-(1-propyn-1-yl)-4-(5-(2,2,2-trifluoro-1-hydroxy-1-methylethyl)-2-pyridinyl)-1-piperazinecarboxylate (1.55 g, 3.46 mmol) was deprotected according to the general Cbz-deprotection method A. The crude product was treated with *tert*-butyl (5-(chlorosulfonyl)-2-pyridinyl)carbamate (**9**) (1.20 g, 4.11 mmol) according to the general sulfonamide formation method using Et_3N (5.0 mL, 36 mmol) and then deprotected according to the general Boc-deprotection method to afford 2-(6-((2*S*)-4-((6-amino-3-pyridinyl)sulfonyl)-2-(1-propyn-1-yl)-1-piperazinyl)-3-pyridinyl)-1,1,1-trifluoro-2-propanol (**28**) (1.42 g, 87% for three steps) as an off-white foam. MS (ESI positive ion) *m/z*: calcd for $C_{20}H_{22}F_3N_5O_3S$, 469.140; found, 470.1 (M + 1). ^{1}H NMR (300 MHz, $CDCl_3$) δ 8.49 (d, *J* = 1.9 Hz, 1 H), 8.38 (s, 1 H), 7.83−7.68 (m, 2 H), 6.67 (s, 1 H), 6.56−6.47 (m, 1 H), 5.22 (br s, 1 H), 4.95 (s, 2 H), 4.11−4.03 (m, 1 H), 3.92−3.77 (m, 2 H), 3.47−3.34 (m, 1 H), 2.70 (dd, *J* = 3.7, 11.3 Hz, 1 H), 2.57 (dt, *J* = 3.2, 11.6 Hz, 1 H), 2.44 (s, 1 H), 1.84−1.72 (m, 6 H).

(2*R*)-2-(6-((2*S*)-4-((6-Amino-3-pyridinyl)sulfonyl)-2-(1-propyn-1-yl)-1-piperazinyl)-3-pyridinyl)-1,1,1-trifluoro-2-propanol (29) and (2*S*)-2-(6-((2*S*)-4-((6-Amino-3-pyridinyl)sulfonyl)-2-(1-propyn-1-yl)-1-piperazinyl)-3-pyridinyl)-1,1,1-trifluoro-2-propanol (30). The individual diastereomers were isolated using chiral SFC from 2-(6-((2*S*)-4-((6-amino-3-pyridinyl)sulfonyl)-2-(1-propyn-1-yl)-1-piperazinyl)-3-pyridinyl)-1,1,1-trifluoro-2-propanol. The method used was as follows: Chiralpak AS-H column (21 mm × 250 mm, 5 μm) using 25% (40 mM NH_3 in methanol) in supercritical CO_2 (total flow was 75 mL/min). This produced the following two diastereomers with diastereomeric and enantiomeric excesses greater than 98%. The absolute stereochemistry around the sulfur in the sulfoximine moiety was determined using enantiomeric pure 2-(6-bromo-3-pyridinyl)-1,1,1-trifluoro-2-propanol.[16]

(2*R*)-2-(6-((2*S*)-4-((6-Amino-3-pyridinyl)sulfonyl)-2-(1-propyn-1-yl)-1-piperazinyl)-3-pyridinyl)-1,1,1-trifluoro-2-propanol (**29**). MS (ESI positive ion) *m/z*: calcd for $C_{20}H_{22}F_3N_5O_3S$, 469.140; found, 470.1 (M + 1). ^{1}H NMR (300 MHz, $CDCl_3$) δ 8.47 (d, *J* = 1.8 Hz, 1 H), 8.37 (d, *J* = 2.3 Hz, 1 H), 7.80−7.67 (m, 2 H), 6.64 (d, *J* = 8.9 Hz, 1 H), 6.51 (d, *J* = 8.8 Hz, 1 H), 5.19 (br s, 1 H), 4.98 (s, 2 H), 4.08 (d, *J* = 12.9 Hz, 1 H), 3.89−3.74 (m, 2 H), 3.39 (dt, *J* = 3.2, 12.4 Hz, 1 H), 2.69 (dd, *J* = 3.5, 11.3 Hz, 1 H), 2.62−2.49 (m, 1 H), 2.40 (br s, 1 H), 1.83−1.68 (m, 6 H).

(2*S*)-2-(6-((2*S*)-4-((6-Amino-3-pyridinyl)sulfonyl)-2-(1-propyn-1-yl)-1-piperazinyl)-3-pyridinyl)-1,1,1-trifluoro-2-propanol (**30**). MS (ESI positive ion) *m/z*: calcd for $C_{20}H_{22}F_3N_5O_3S$, 469.140; found, 470.1 (M + 1). ^{1}H NMR (300 MHz, $CDCl_3$) δ 8.47 (d, *J* = 1.9 Hz, 1 H), 8.36 (d, *J* = 2.5 Hz, 1 H), 7.82−7.66 (m, 2 H), 6.64 (d, *J* = 8.9 Hz, 1 H), 6.50 (d, *J* = 8.8 Hz, 1 H), 5.20 (br s, 1 H), 4.97 (s, 2 H), 4.07 (d, *J* = 13.4 Hz, 1 H), 3.91−3.73 (m, 2 H), 3.39 (dt, *J* = 3.1, 12.3 Hz, 1 H), 2.69 (dd, *J* = 3.5, 11.3 Hz, 1 H), 2.55 (dt, *J* = 3.3, 11.7 Hz, 1 H), 2.39 (br s, 1 H), 1.83−1.69 (m, 6 H).

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5-(((3*S*)-4-(4-(*S*-Cyclopropylsulfonimidoyl)phenyl)-3-(1-propyn-1-yl)-1-piperazinyl)sulfonyl)-2-pyridinamine (47). (3*S*)-1-Benzyl-3-(1-propyn-1-yl)piperazine (**5**) (0.510 g, 2.41 mmol) and 1-bromo-4-(cyclopropylsulfanyl)benzene[13] (0.500 g, 2.19 mmol) were coupled according to the general amination method using RuPhos (0.051 g, 0.109 mmol), $Pd_2(dba)_3$ (0.060 g, 0.066 mmol), and 1,4-dioxane to give (2*S*)-4-benzyl-1-(4-(cyclopropylsulfanyl)phenyl)-2-(1-propyn-1-yl)piperazine (**31**) (0.350 g, 47%) as a white solid. MS (ESI positive ion) *m*/*z*: calcd for $C_{23}H_{26}N_2S$, 362.182; found, 363.2 (M + 1).

Compound **31** (0.350 g, 0.690 mmol) was deprotected according to the general Bn-deprotection method to afford (2*S*)-1-(4-(cyclopropylsulfanyl)phenyl)-2-(1-propyn-1-yl)piperazine (**32**) (0.40 g, crude) as a pale-brown residue, which was carried forward to the next step without further purification. MS (ESI positive ion) *m*/*z*: calcd for $C_{16}H_{20}N_2S$, 272.135; found, 273.1 (M + 1).

Compound **32** (0.400 g, 1.47 mmol) was treated with *tert*-butyl (5-(chlorosulfonyl)-2-pyridinyl)carbamate (**9**) (0.510 g, 1.76 mmol) according to the general sulfonamide formation method using pyridine (2.0 mL, 25 mmol) to give *tert*-butyl (5-(((3*S*)-4-(4-(cyclopropylsulfanyl)phenyl)-3-(1-propyn-1-yl)-1-piperazinyl)-sulfonyl)-2-pyridinyl)carbamate (**41**) (0.260 g, 50% over two steps) as a white solid. MS (ESI positive ion) *m*/*z*: calcd for $C_{26}H_{32}N_4O_4S_2$, 528.186; found, 528.7 (M + 1). ^{1}H NMR (300 MHz, DMSO-d_6) δ 10.50 (s, 1 H), 8.59 (d, *J* = 2.4 Hz, 1 H), 8.15−7.98 (m, 2 H), 7.25 (d, *J* = 8.3 Hz, 2 H), 6.93 (d, *J* = 8.4 Hz, 2 H), 4.70 (s, 1 H), 3.66 (m, 2 H), 3.42 (d, *J* = 12.1 Hz, 2 H), 3.17−2.96 (m, 1 H), 2.71−2.58 (m, 1 H), 2.22 (m, 1 H), 1.74 (d, *J* = 2.0 Hz, 3 H), 1.49 (d, *J* = 4.1 Hz, 9 H), 0.99 (td, *J* = 6.7, 4.4 Hz, 2 H), 0.58−0.49 (m, 2 H).

A 100 mL round-bottomed flask was charged with compound **41** (0.26 g, 0.49 mmol) and acetic acid (2.6 mL). The mixture was cooled to 0 °C, and H_2O_2 (30%, 0.075 g, 0.74 mmol) was added dropwise over a period of 5 min. The resulting reaction mixture was stirred at 0 °C for 30 min and at room temperature for 1 h. The reaction mixture was diluted with saturated aqueous $NaHCO_3$ solution (30 mL) and EtOAc (30 mL). The organic layer was separated, washed with water and saturated aqueous NaCl, dried over anhydrous Na_2SO_4, and filtered. The filtrate was concentrated under reduced pressure and the residue was purified by silica gel column chromatography (eluent, 70% EtOAc−hexanes) to give *tert*-butyl (5-(((3*S*)-4-(4-(cyclopropylsulfinyl)phenyl)-3-(1-propyn-1-yl)-1-piperazinyl)-sulfonyl)-2-pyridinyl)carbamate (**42**) (0.150 g, 56%) as a pale-brown viscous solid. MS (ESI positive ion) *m*/*z*: calcd for $C_{26}H_{32}N_4O_5S_2$, 544.181; found, 545.0 (M + 1).

A 100 mL round-bottomed flask was charged with compound **42** (0.1 g, 0.2 mmol), $CHCl_3$ (2 mL), and NaN_3 (0.020 g, 0.29 mmol). The resulting mixture was cooled to 0 °C, and concentrated H_2SO_4 (0.095 g, 0.97 mmol) was added dropwise. The reaction mixture was allowed to warm to room temperature and stirred at room temperature for 6 h. The reaction mixture was diluted with ice-cold water (20 mL) and neutralized with Na_2CO_3 solution (10%, 10 mL) before extracting with EtOAc (3 × 50 mL). The combined organic extracts were washed with water and saturated aqueous NaCl, dried over anhydrous Na_2SO_4, filtered, and concentrated under reduced pressure. The residue obtained was purified by silica gel column chromatography (eluent, 10% MeOH−DCM) to give 5-(((3*S*)-4-(4-(*S*-cyclopropylsulfonimidoyl)phenyl)-3-(1-propyn-1-yl)-1-piperazinyl)-sulfonyl)-2-pyridinamine (**47**) (0.010 g, 11%). MS (ESI positive ion) *m*/*z*: calcd for $C_{21}H_{22}N_5O_3S_2$, 459.140; found, 459.9 (M + 1). ^{1}H NMR (300 MHz, CD_3OD) δ 8.20 (d, *J* = 2.4 Hz, 1 H), 7.72−7.57 (m, 3 H), 7.01 (d, *J* = 8.8 Hz, 2 H), 6.52 (d, *J* = 9.0 Hz, 1 H), 4.71 (s, 1 H), 3.67 (bd, *J* = 11.2 Hz, 2 H), 3.57 (d, *J* = 12.4 Hz, 1 H), 2.69−2.38 (m, 3 H), 1.66 (dd, *J* = 2.2, 0.9 Hz, 3 H), 1.13 (q, *J* = 6.5 Hz, 2 H), 0.96 (tt, *J* = 9.5, 4.7 Hz, 2 H). One proton is obscured under the solvent peak. HPLC purity at 254 nm, 90.3%; at 215 nm, 91.5%.

5-(((3*S*)-4-(4-(*S*-Methylsulfonimidoyl)phenyl)-3-(1-propyn-1-yl)-1-piperazinyl)sulfonyl)-2-pyridinamine (48). Benzyl (3*S*)-3-(1-propyn-1-yl)-1-piperazinecarboxylate (**4**) (0.50 g, 1.9 mmol) and 1-bromo-4-(methylsulfinyl)benzene[13] (0.63 g, 2.9 mmol) were coupled according to the general amination method using $Pd_2(dba)_3$ (0.089 g, 0.096 mmol), DavePhos (0.037 g, 0.096 mmol), and toluene to afford benzyl (3*S*)-4-(4-(methylsulfinyl)phenyl)-3-(1-propyn-1-yl)-1-piperazinecarboxylate (**33**) (0.75 g, 65%) as a pale-brown gummy solid. MS (ESI positive ion) *m*/*z*: calcd for $C_{22}H_{24}N_2O_3S$, 396.151; found, 397.0 (M + 1).

Compound **33** (0.75 g, 1.9 mmol) was deprotected according to the general Cbz-deprotection method B to give (2*S*)-1-(4-(methylsulfinyl)phenyl)-2-(1-propyn-1-yl)piperazine (**34**) (0.40 g, 80%) as a brown gummy liquid. MS (ESI positive ion) *m*/*z*: calcd for $C_{14}H_{18}N_2OS$, 262.114; found, 263.1 (M + 1).

Compound **34** (0.40 g, 1.5 mmol) was treated with *tert*-butyl (5-(chlorosulfonyl)-2-pyridinyl)carbamate (**9**) (0.67 g, 2.3 mmol) according to the general sulfonamide formation method using Et_3N (0.6 mL, 3.8 mmol) to give *tert*-butyl (5-(((3*S*)-4-(4-(methylsulfinyl)-phenyl)-3-(1-propyn-1-yl)-1-piperazinyl)sulfonyl)-2-pyridinyl)-carbamate (**43**) (0.35 g, 44%) as a brown solid. MS (ESI positive ion) *m*/*z*: calcd for $C_{24}H_{30}N_4O_5S_2$, 518.166; found, 519.3 (M + 1). ^{1}H NMR (300 MHz, $CDCl_3$) δ 8.66 (s, 1 H), 8.13 (d, *J* = 6.6 Hz, 1 H), 8.03 (dd, *J* = 6.6, 1.5 Hz, 1 H), 7.65 (s, 1 H), 7.56 (d, *J* = 6.6 Hz, 2 H), 7.03 (d, *J*= 6.0 Hz, 2 H), 4.47 (s, 1 H), 3.84−3.77 (m, 2 H), 3.43−3.39 77 (m, 3 H), 2.84 (dd, *J* = 8.4, 2.7 Hz, 1 H), 2.70 (s, 3 H), 1.78 (s, 3 H), 1.58 (s, 9 H).

A 100 mL round-bottomed flask was charged with compound **43** (0.25 g, 0.48 mmol) and $CHCl_3$ (20 mL) and treated with NaN_3 (0.035 g, 0.53 mmol) at room temperature. The resulting mixture was cooled to 0 °C, and concentrated H_2SO_4 (0.530 g, 5.3 mmol) was added dropwise to the above solution. The reaction mixture was allowed to warm to room temperature and stirred at room temperature for 6 h. The reaction mixture was diluted with ice-cold water (20 mL) and neutralized with Na_2CO_3 solution (10%, 10 mL) before extracting with $CHCl_3$ (3 × 50 mL). The combined organic extracts were washed with water and saturated aqueous NaCl, dried over anhydrous Na_2SO_4, filtered, and concentrated under reduced pressure. The residue obtained was purified by silica gel preparative TLC (eluent 3% MeOH−DCM) to give 5-(((3*S*)-4-(4-(*S*-methylsulfonimidoyl)-phenyl)-3-(1-propyn-1-yl)-1-piperazinyl)sulfonyl)-2-pyridinamine (**48**) (0.075 g, 36%) as a white solid. MS (ESI positive ion) *m*/*z*: calcd for $C_{19}H_{23}N_5O_3S_2$, 433.124; found, 434.1 (M + 1). ^{1}H NMR (400 MHz, DMSO-d_6) δ 8.24 (d, *J* = 2.4 Hz 1 H), 7.74 (d, *J* = 8.8 Hz, 2 H), 7.64 (dd, *J* = 8.8, 2.4 Hz, 1H), 7.08 (d, *J* = 8.8 Hz, 2 H), 7.04 (s, 2 H), 6.53 (d, *J* = 8.8 Hz, 1 H), 4.93 (s, 1 H), 3.96 (d, *J* = 4.8 Hz, 1 H), 3.72−3.60 (m, 3 H), 3.17−3.12 (m, 2 H), 2.99 (s, 3 H), 2.40−2.33 (m, 1 H), 1.76 (d, *J* = 1.6 Hz, 3 H).

4-((2*S*)-4-((6-Amino-3-pyridinyl)sulfonyl)-2-(1-propyn-1-yl)-1-piperazinyl)benzenesulfonamide (49). Benzyl (3*S*)-3-(1-propyn-1-yl)-1-piperazinecarboxylate (**4**) (0.440 g, 1.73 mmol) and 4-bromo-*N*-(cyclopropylmethyl)benzenesulfonamide[13] (0.500 g, 1.73 mmol) were coupled according to the general amination method using JohnPhos (0.025 g, 0.086 mmol), $Pd_2(dba)_3$ (0.047 g, 0.051 mmol), and 1,4-dioxane to obtain benzyl (3*S*)-4-(4-((cyclopropylmethyl)-sulfamoyl)phenyl)-3-(1-propyn-1-yl)-1-piperazinecarboxylate (**35**) (0.270 g, 33%) as a white solid. MS (ESI positive ion) *m*/*z*: calcd for $C_{25}H_{29}N_3O_4S$, 467.188; found, 468.0 (M + 1).

Compound **35** (0.270 g, 0.58 mmol) was globally deprotected according to the general Cbz-deprotection method B to give 4-((2*S*)-2-(1-propyn-1-yl)-1-piperazinyl)benzenesulfonamide (**36**) as a pale-brown solid (0.19 g, crude), which was used in the next step without purification.

Compound **36** (0.100 g, 0.35 mmol) was treated with *tert*-butyl (5-(chlorosulfonyl)-2-pyridinyl)carbamate (**9**) (0.156 g, 0.53 mmol) according to the general sulfonamide formation method using pyridine (1.0 mL, 12 mmol) to afford *tert*-butyl (5-(((3*S*)-3-(1-propyn-1-yl)-4-(4-sulfamoylphenyl)-1-piperazinyl)sulfonyl)-2-pyridinyl)carbamate (**44**) (0.060 g, 28% over two steps) as a white solid. MS (ESI positive ion) *m*/*z*: calcd for $C_{23}H_{29}N_5O_6S_2$, 535.156; found, 558.1 (M + Na). ^{1}H NMR (300 MHz, $CDCl_3$) δ 8.66 (d, *J* = 2.4 Hz, 1 H), 8.14 (d, *J* = 9 Hz, 1 H), 8.03 (dd, *J* = 9.0, 2.4 Hz, 1 H), 7.80 (d, *J* = 9 Hz, 3 H), 6.95 (d, *J* = 9 Hz, 2 H), 4.74 (s, 2 H), 4.52 (s, 1 H), 3.91−3.75 (m, 2 H), 3.57−3.33 (m, 2 H), 2.81 (dd, *J* = 11.4, 3.5 Hz, 1 H), 2.67 (td, *J* = 11.4, 3.6 Hz, 1 H), 1.78 (d, *J* = 2.1 Hz, 3 H), 1.54 (s, 9 H).

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Compound **44** was deprotected according to the general Boc-deprotection method to afford 4-((2*S*)-4-((6-amino-3-pyridinyl)-sulfonyl)-2-(1-propyn-1-yl)-1-piperazinyl)benzenesulfonamide (**49**) (0.010 g, 12%) as a white solid. MS (ESI positive ion) *m*/*z*: calcd for $C_{18}H_{21}N_5O_4S_2$, 435.103; found, 436.1 (M + 1). ^{1}H NMR (400 MHz, DMSO-d_6) δ 8.26 (d, *J* = 2.5 Hz, 1 H), 7.70−7.63 (m, 3 H), 7.16 (s, 2 H), 7.08 (d, *J* = 8.8 Hz, 2 H), 6.56 (d, *J* = 8.9 Hz, 1 H), 4.94 (s, 1 H), 3.72−3.59 (m, 4 H), 3.17−3.04 (m, 1 H), 2.45−2.32 (m, 2 H), 1.77 (d, *J* = 2.1 Hz, 3 H). One proton was obscured under the solvent peak.

(2*S*)-2-(4-((2*S*)-4-((6-Amino-3-pyridinyl)sulfonyl)-2-(1-propyn-1-yl)-1-piperazinyl)phenyl)-3,3,3-trifluoro-1,2-propanediol (51) and (2*R*)-2-(4-((2*S*)-4-((6-Amino-3-pyridinyl)sulfonyl)-2-(1-propyn-1-yl)-1-piperazinyl)phenyl)-3,3,3-trifluoro-1,2-propanediol (52). Benzyl (3*S*)-3-(1-propyn-1-yl)-1-piperazinecarboxylate (**4**) (5.0 g, 19 mmol) and 4-(4-bromophenyl)-2,2-dimethyl-4-(trifluoromethyl)-1,3-dioxolane[13] (6.0 g, 20 mmol) were coupled according to the general amination method using RuPhos (0.45 g, 0.97 mmol), RuPhos first generation precatalyst (0.79 g, 0.97 mmol), and 1,4-dioxane to afford benzyl (3*S*)-4-(4-(2,2-dimethyl-4-(trifluoromethyl)-1,3-dioxolan-4-yl)phenyl)-3-(1-propyn-1-yl)-1-piperazinecarboxylate (**37**) (5.0 g, 51%). MS (ESI positive ion) *m*/*z*: calcd for $C_{27}H_{29}F_3N_2O_4$, 502.208; found, 503.2 (M + 1). ^{1}H NMR (300 MHz, $CDCl_3$) δ 7.44−7.31 (m, 7 H), 6.96 (d, *J* = 8.6 Hz, 2 H), 5.35−5.09 (m, 2 H), 4.74−4.63 (m, 1 H), 4.43−4.18 (m, 4 H), 3.44−3.21 (m, 3 H), 3.12 (d, *J* = 13.2 Hz, 1 H), 1.77−1.68 (m, 3 H), 1.63−1.58 (m, 3 H), 1.37−1.30 (m, 3 H).

Compound **37** (5.0 g, 9.9 mmol) was globally deprotected according to the general Cbz-deprotection method A. The crude product obtained was treated with *tert*-butyl (5-(chlorosulfonyl)-2-pyridinyl)carbamate (**9**) (3.50 g, 12.0 mmol) according to the general sulfonamide formation using Et_3N (7.42 mL, 53.4 mmol) and then deprotected according to the general Boc-deprotection method to afford 2-(4-((2*S*)-4-((6-amino-3-pyridinyl)sulfonyl)-2-(1-propyn-1-yl)-1-piperazinyl)phenyl)-3,3,3-trifluoro-1,2-propanediol (**50**) (4.7 g, 91% over three steps) as a light-brown foam. MS (ESI positive ion) *m*/*z*: calcd for $C_{21}H_{23}F_3N_4O_4S$, 484.139; found, 485.1 (M + 1). ^{1}H NMR (300 MHz, $CDCl_3$) δ 8.50 (d, *J* = 2.3 Hz, 1 H), 7.78 (dd, *J* = 2.4, 8.7 Hz, 1 H), 7.45 (d, *J* = 8.6 Hz, 2 H), 6.96 (d, *J* = 8.9 Hz, 2 H), 6.53 (d, *J* = 8.8 Hz, 1 H), 4.97 (s, 2 H), 4.41 (br s, 1 H), 4.26 (d, *J* = 9.2 Hz, 1 H), 3.90 (d, *J* = 12.9 Hz, 1 H), 3.81−3.59 (m, 3 H), 3.45−3.30 (m, 2 H), 2.85 (dd, *J* = 3.4, 11.1 Hz, 1 H), 2.70 (td, *J* = 7.3, 11.1 Hz, 1 H), 1.92 (br s, 1 H), 1.79 (d, *J* = 1.9 Hz, 3 H).

The diastereomeric mixture **50** was resolved using preparative SFC (Chiralpak AS-H (250 mm × 21 mm, 5 μm, using 30% (20 mM NH_3 in methanol) in super critical CO_2 (total flow was 75 mL/min). This produced the following two compounds with diastereomeric excess values greater than 99%, each. The absolute chemistry of each compound was established by X-ray crystallography of cocrystals of these molecules to hGKRP protein.

(2*S*)-2-(4-((2*S*)-4-((6-Amino-3-pyridinyl)sulfonyl)-2-(1-propyn-1-yl)-1-piperazinyl)phenyl)-3,3,3-trifluoro-1,2-propanediol (**51**). MS (ESI positive ion) *m*/*z*: calcd for $C_{21}H_{23}F_3N_4O_4S$, 484.139; found, 485.1 (M + 1). ^{1}H NMR (300 MHz, $CDCl_3$) δ 8.48 (d, *J* = 2.2 Hz, 1 H), 7.77 (dd, *J* = 2.3, 8.8 Hz, 1 H), 7.44 (d, *J* = 8.8 Hz, 2 H), 6.95 (d, *J* = 8.9 Hz, 2 H), 6.52 (d, *J* = 8.6 Hz, 1 H), 4.98 (s, 2 H), 4.41 (d, *J* = 2.0 Hz, 1 H), 4.25 (d, *J* = 11.8 Hz, 1 H), 3.89 (d, *J* = 11.3 Hz, 1 H), 3.80−3.58 (m, 3 H), 3.43−3.34 (m, *J* = 2.8 Hz, 2 H), 2.84 (dd, *J* = 3.4, 11.2 Hz, 1 H), 2.74−2.63 (m, 1 H), 1.77 (d, *J* = 2.0 Hz, 3 H). One exchangeable proton was not observed.

(2*R*)-2-(4-((2*S*)-4-((6-Amino-3-pyridinyl)sulfonyl)-2-(1-propyn-1-yl)-1-piperazinyl)phenyl)-3,3,3-trifluoro-1,2-propanediol (**52**). MS (ESI positive ion) *m*/*z*: calcd for $C_{21}H_{23}F_3N_4O_4S$, 484.139; found, 485.1 (M + 1). ^{1}H NMR (300 MHz, $CDCl_3$) δ 8.50 (d, *J* = 2.2 Hz, 1 H), 7.78 (dd, *J* = 2.4, 8.8 Hz, 1 H), 7.45 (d, *J* = 8.6 Hz, 2 H), 6.96 (d, *J* = 8.9 Hz, 2 H), 6.54 (d, *J* = 8.8 Hz, 1 H), 4.98 (s, 2 H), 4.41 (br s, 1 H), 4.26 (d, *J* = 11.8 Hz, 1 H), 3.90 (d, *J* = 12.1 Hz, 1 H), 3.75 (t, *J* = 10.1 Hz, 2 H), 3.64 (br s, 1 H), 3.44−3.32 (m, 2 H), 2.85 (dd, *J* = 3.1, 11.1 Hz, 1 H), 2.77−2.60 (m, 1 H), 1.79 (d, *J* = 1.9 Hz, 3 H). One exchangeable proton was not observed.

2-(6-((2*S*)-4-((6-Amino-3-pyridinyl)sulfonyl)-2-(1-propyn-1-yl)-1-piperazinyl)-3-pyridinyl)-3,3,3-trifluoro-1,2-propanediol (53). Benzyl (3*S*)-3-(1-propyn-1-yl)-1-piperazinecarboxylate (**4**) (0.100 g, 0.387 mmol) and 2-bromo-5-(2,2-dimethyl-4-(trifluoromethyl)-1,3-dioxolan-4-yl)pyridine[13](0.140 g, 0.429 mmol) were coupled according to the general amination method using RuPhos (0.009 g, 0.020 mmol), RuPhos first generation precatalyst (0.016 g, 0.019 mmol), and 1,4-dioxane to afford benzyl (3*S*)-4-(5-(2,2-dimethyl-4-(trifluoromethyl)-1,3-dioxolan-4-yl)-2-pyridinyl)-3-(1-propyn-1-yl)-1-piperazinecarboxylate (**39**) (0.045 g, 23%) as a yellow paste. MS (ESI positive ion) *m*/*z*: calcd for $C_{26}H_{28}F_3N_3O_4S$, 503.203; found, 504.2 (M + 1). ^{1}H NMR (300 MHz, $CDCl_3$) δ 8.29 (s, 1 H), 7.61 (d, *J* = 9.1 Hz, 1 H), 7.45−7.30 (m, 5 H), 6.68 (d, *J* = 8.9 Hz, 1 H), 5.28−5.08 (m, 3 H), 4.73−4.62 (m, 1 H), 4.35 (br s, 2 H), 4.26−4.17 (m, 1 H), 3.99 (br s, 1 H), 3.34 (t, *J* = 11.5 Hz, 1 H), 3.21 (br s, 1 H), 3.06 (d, *J* = 14.2 Hz, 1 H), 1.71 (br s, 3 H), 1.63−1.57 (m, 3 H), 1.34 (s, 3 H).

Compound **39** (0.100 g, 0.199 mmol) was globally deprotected according to the general Cbz-deprotection method A. The crude product was treated with *tert*-butyl (5-(chlorosulfonyl)-2-pyridinyl)-carbamate (**9**) (0.065 g, 0.222 mmol) according to the general sulfonamide formation method using Et_3N (0.14 mL, 0.99 mmol) and then deprotected according to the general Boc-deprotection method to afford 2-(6-((2*S*)-4-((6-amino-3-pyridinyl)sulfonyl)-2-(1-propyn-1-yl)-1-piperazinyl)-3-pyridinyl)-3,3,3-trifluoro-1,2-propanediol (**53**) (0.060 g, 62% over three steps) as a light-brown foam. MS (ESI positive ion) *m*/*z*: calcd for $C_{20}H_{22}F_3N_5O_4S$, 485.134; found, 508.1 (M + Na). ^{1}H NMR (300 MHz, $CDCl_3$) δ 8.48 (d, *J* = 2.2 Hz, 1 H), 8.34 (br s, 1 H), 7.86−7.63 (m, 2 H), 6.67 (d, *J* = 8.9 Hz, 1 H), 6.52 (d, *J* = 8.8 Hz, 1 H), 5.20 (br s, 1 H), 4.96 (s, 2 H), 4.28 (d, *J* = 11.8 Hz, 1 H), 4.10 (d, *J* = 12.4 Hz, 1 H), 3.96−3.75 (m, 3 H), 3.67 (br s, 1 H), 3.49−3.28 (m, 1 H), 2.72 (d, *J* = 8.2 Hz, 1 H), 2.66−2.45 (m, 1 H), 1.80 (d, *J* = 1.8 Hz, 3 H). One exchangeable proton was not observed.

(2*S*)-2-(6-((2*S*)-4-((6-Amino-3-pyridinyl)sulfonyl)-2-(1-propyn-1-yl)-1-piperazinyl)-3-pyridinyl)-3,3,3-trifluoro-1,2-propanediol (54) and (2*R*)-2-(6-((2*S*)-4-((6-Amino-3-pyridinyl)-sulfonyl)-2-(1-propyn-1-yl)-1-piperazinyl)-3-pyridinyl)-3,3,3-trifluoro-1,2-propanediol (55). The diastereomeric mixture of 2-(6-((2*S*)-4-((6-amino-3-pyridinyl)sulfonyl)-2-(1-propyn-1-yl)-1-piperazinyl)-3-pyridinyl)-3,3,3-trifluoro-1,2-propanediol (**53**) was separated by chiral SFC method (ODH (21 mm × 250 mm, 5 μm), 20% MeOH with 0.2% diethylamine in CO_2, flow rate of 70 mL/min) to afford each diastereomer. The stereochemistry at the carbinol center was assigned arbitrarily.

(2*S*)-2-(6-((2*S*)-4-((6-Amino-3-pyridinyl)sulfonyl)-2-(1-propyn-1-yl)-1-piperazinyl)-3-pyridinyl)-3,3,3-trifluoro-1,2-propanediol (**54**). MS (ESI positive ion) *m*/*z*: calcd for $C_{20}H_{22}F_3N_5O_4S$, 485.134; found, 508.1 (M + Na). ^{1}H NMR (300 MHz, $CDCl_3$) δ 8.48 (d, *J* = 1.9 Hz, 1 H), 8.34 (s, 1 H), 7.83−7.66 (m, 2 H), 6.67 (d, *J* = 9.1 Hz, 1 H), 6.52 (d, *J* = 8.9 Hz, 1 H), 5.20 (br s, 1 H), 4.97 (s, 2 H), 4.28 (d, *J* = 12.0 Hz, 1 H), 4.10 (d, *J* = 13.6 Hz, 1 H), 3.94−3.76 (m, 3 H), 3.67 (br s, 1 H), 3.53−3.34 (m, 1 H), 2.73 (dd, *J* = 3.7, 11.4 Hz, 1 H), 2.66−2.50 (m, 1 H), 1.80 (d, *J* = 2.0 Hz, 3 H). One exchangeable proton was not observed.

(2*R*)-2-(6-((2*S*)-4-((6-Amino-3-pyridinyl)sulfonyl)-2-(1-propyn-1-yl)-1-piperazinyl)-3-pyridinyl)-3,3,3-trifluoro-1,2-propanediol (**55**). MS (ESI positive ion) *m*/*z*: calcd for $C_{20}H_{22}F_3N_5O_4S$, 485.134; found, 508.1 (M + Na). ^{1}H NMR (300 MHz, DMSO-d_6) δ 8.28 (m, 1 H), 8.23 (d, *J* = 2.2 Hz, 1 H), 7.73 (dd, *J* = 2.2, 8.9 Hz, 1 H), 7.63 (dd, *J* = 2.5, 8.9 Hz, 1 H), 6.99 (s, 2 H), 6.84 (d, *J* = 9.1 Hz, 1 H), 6.52 (d, *J* = 8.9 Hz, 1 H), 6.43 (s, 1 H), 5.34 (br s, 1 H), 5.16 (t, *J* = 5.8 Hz, 1 H), 4.15 (d, *J* = 12.6 Hz, 1 H), 3.97−3.76 (m, 2 H), 3.64 (d, *J* = 10.1 Hz, 2 H), 3.14−3.05 (m, 1 H), 2.45 (d, *J* = 3.1 Hz, 1 H), 2.37−2.21 (m, 1 H), 1.75 (d, *J* = 2.0 Hz, 3 H).

3-(4-((2*S*)-4-((6-Amino-3-pyridinyl)sulfonyl)-2-(1-propyn-1-yl)-1-piperazinyl)phenyl)-4,4,4-trifluoro-3-hydroxybutanenitrile (62). Compound **37** (1.60 g, 3.18 mmol) was globally deprotected according to the general Cbz-deprotection method. The crude product was treated with *tert*-butyl (5-(chlorosulfonyl)-2-pyridinyl)carbamate (**9**) (1.04 g, 3.60 mmol) according to the general sulfonamide formation method using Et_3N (2.20 mL, 16.0 mmol) to

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afford *tert*-butyl(5-(((3*S*)-3-(1-propyn-1-yl)-4-(4-(2,2,2-trifluoro-1-hydroxy-1-(hydroxymethyl)ethyl)phenyl)-1-piperazinyl)sulfonyl)-2-pyridinyl)carbamate (**45**) (1.0 g, 54%) as a yellow foam. MS (ESI positive ion) *m*/*z*: calcd for $C_{26}H_{31}F_3N_4O_6S$, 584.192; found, 585.2 (M + 1). ^{1}H NMR (300 MHz, $CDCl_3$) δ 8.67 (d, *J* = 2.3 Hz, 1 H), 8.15 (d, *J* = 8.8 Hz, 1 H), 8.04 (dd, *J* = 2.3, 8.8 Hz, 1 H), 7.94 (s, 1 H), 7.45 (d, *J* = 8.6 Hz, 2 H), 6.95 (d, *J* = 8.8 Hz, 2 H), 4.42 (br s, 1 H), 4.26 (dd, *J* = 6.5, 11.9 Hz, 1 H), 3.95−3.85 (m, 1 H), 3.84−3.69 (m, 2 H), 3.63 (s, 1 H), 3.43−3.33 (m, 2 H), 2.85 (dd, *J* = 3.3, 11.2 Hz, 1 H), 2.77−2.63 (m, 1 H), 1.86 (dt, *J* = 2.4, 6.8 Hz, 1 H), 1.78 (d, *J* = 2.0 Hz, 3 H), 1.56 (s, 9 H).

To a solution of compound **45** (0.300 g, 0.513 mmol) in DCM (5 mL) were added Et_3N (0.400 mL, 2.88 mmol) and *p*-toluenesulfonyl chloride (0.108 g, 0.564 mmol). The resulting mixture was heated at reflux (50 °C) under N_2 for 2 h. The reaction mixture was allowed to cool to room temperature and partitioned between saturated $NaHCO_3$ (30 mL) and DCM (70 mL). The aqueous layer was extracted with DCM (2 × 40 mL). The combined organic layers were dried over $MgSO_4$, filtered, and concentrated. The crude product was purified by silica gel column chromatography (10−40% acetone in hexanes) to afford *tert*-butyl (5-(((3*S*)-3-(1-propyn-1-yl)-4-(4-(2-(trifluoromethyl)-2-oxiranyl)phenyl)-1-piperazinyl)sulfonyl)-2-pyridinyl)carbamate (**56**) (0.240 g, 83%) as an off-white solid. MS (ESI positive ion) *m*/*z*: calcd for $C_{26}H_{29}F_3N_4O_5S$, 566.181; found, 567.2 (M + 1). ^{1}H NMR (400 MHz, $CDCl_3$) δ 8.66 (dd, *J* = 0.6, 2.3 Hz, 1 H), 8.20−8.10 (m, 1 H), 8.04 (dd, *J* = 2.2, 8.9 Hz, 1 H), 7.63 (s, 1 H), 7.41 (d, *J* = 8.6 Hz, 2 H), 6.94 (d, *J* = 8.8 Hz, 2 H), 4.42 (d, *J* = 2.2 Hz, 1 H), 3.89−3.67 (m, 2 H), 3.38 (d, *J* = 5.3 Hz, 3 H), 2.97−2.83 (m, 2 H), 2.80−2.60 (m, 1 H), 1.78 (dd, *J* = 0.8, 2.0 Hz, 3 H), 1.55 (s, 9 H).

To a solution of compound **56** (0.030 g, 0.053 mmol) in DMF (2 mL) were added water (0.3 mL) and KCN (10 mg, 0.154 mmol). The mixture was stirred at room temperature for 20 min and then partitioned between EtOAc (30 mL) and water (10 mL). The organic layer was washed with water (3×), saturated NaCl, dried over Na_2SO_4, filtered, and concentrated to give a crystalline solid. This solid was dissolved in DCM (2.5 mL) and was treated with TFA (0.300 mL, 3.89 mmol). After 1 h, the mixture was concentrated and the residue was diluted with EtOAc (20 mL) and washed with 1 N aqueous NaOH (5 mL), water (5 mL), and saturated NaCl (5 mL). The organic layer was dried over Na_2SO_4, filtered, and concentrated. The residue was purified by chromatography on silica gel using EtOAc in hexanes (30−80%) as eluent to give 3-(4-((2*S*)-4-((6-amino-3-pyridinyl)sulfonyl)-2-(1-propyn-1-yl)-1-piperazinyl)phenyl)-4,4,4-trifluoro-3-hydroxybutanenitrile (**62**) (0.020 g, 77%) as a light-pink foam. MS (ESI positive ion) *m*/*z*: calcd for $C_{22}H_{22}F_3N_5O_3S$, 493.140; found, 494.3 (M + 1). ^{1}H NMR (400 MHz, $CDCl_3$) δ 8.48 (br s, 1 H), 7.79 (d, *J* = 8.8 Hz, 1 H), 7.44 (d, *J* = 7.6 Hz, 2 H), 6.97 (d, *J* = 7.4 Hz, 2 H), 6.54 (d, *J* = 8.2 Hz, 1 H), 5.10 (br s, 2 H), 4.44 (br s, 1 H), 4.07−4.18 (m, 1 H), 3.34−3.49 (m, 2 H), 3.17 (br s, 2 H), 2.95−3.07 (m, 1 H), 2.85 (d, *J* = 11.0 Hz, 1 H), 2.70 (t, *J* = 13.1 Hz, 1 H), 1.79 (br s, 3 H). One exchangeable proton was not observed.

2-(4-((2*S*)-4-((6-Amino-3-pyridinyl)sulfonyl)-2-(1-propyn-1-yl)-1-piperazinyl)phenyl)-1,1,1-trifluoro-3-methoxy-2-propanol (63). A flame-dried 50 mL two-necked round-bottomed flask was charged with sodium methoxide (0.045 g, 0.85 mmol) and anhydrous MeOH (3 mL). To this solution was added the suspension of *tert*-butyl (5-(((3*S*)-3-(1-propyn-1-yl)-4-(4-(2-(trifluoromethyl)-2-oxiranyl)phenyl)-1-piperazinyl)sulfonyl)-2-pyridinyl)carbamate (**56**) (0.080 g, 0.14 mmol) in MeOH (3 mL). The resulting mixture was heated at 70 °C for 1 h, and 3 mL of DMF was added. The stirring at 70 °C was resumed for 24 h. The reaction mixture was allowed to cool to room temperature and concentrated. The residue was partitioned between EtOAc (60 mL) and water (30 mL), and the aqueous layer was extracted with EtOAc (30 mL). The combined organic layers were dried over $MgSO_4$, filtered, and concentrated. The crude product was purified by silica gel column chromatography (10−40% EtOAc in hexanes) to afford the crude *tert*-butyl (5-(((3*S*)-3-(1-propyn-1-yl)-4-(4-(2,2,2-trifluoro-1-hydroxy-1-(methoxymethyl)ethyl)phenyl)-1-piperazinyl)sulfonyl)-2-pyridinyl)carbamate (**58**) (0.045 g). This material was dissolved in DCM (3.0 mL) and treated with TFA (0.024 mL, 0.33 mmol). The mixture was stirred at room temperature for 1 h. The reaction mixture was partitioned between saturated $NaHCO_3$ (15 mL) and DCM (40 mL), and the aqueous layer was extracted with DCM (2 × 30 mL). The combined organic layers were dried over $MgSO_4$, filtered, and concentrated. The crude product was purified by silica column chromatography (0−5% MeOH in DCM) to afford 2-(4-((2*S*)-4-((6-amino-3-pyridinyl)sulfonyl)-2-(1-propyn-1-yl)-1-piperazinyl)phenyl)-1,1,1-trifluoro-3-methoxy-2-propanol (**63**) (0.030 g, 43% over two steps) as an off-white solid. MS (ESI positive ion) *m*/*z*: calcd for $C_{22}H_{25}F_3N_4O_4S$, 498.155; found, 499.1 (M + 1). ^{1}H NMR (300 MHz, $CDCl_3$) δ 8.49 (d, *J* = 2.0 Hz, 1 H), 7.82 (dd, *J* = 2.4, 8.8 Hz, 1 H), 7.44 (d, *J* = 8.8 Hz, 2 H), 6.95 (d, *J* = 8.9 Hz, 2 H), 6.59 (d, *J* = 8.9 Hz, 1 H), 5.27 (br s, 2 H), 4.42 (br s, 1 H), 4.02 (d, *J* = 9.9 Hz, 1 H), 3.82−3.68 (m, 2 H), 3.69−3.57 (m, 2 H), 3.45 (s, 3 H), 3.41−3.33 (m, 2 H), 2.87 (dd, *J* = 3.4, 11.1 Hz, 1 H), 2.72 (td, *J* = 7.1, 11.4 Hz, 1 H), 1.79 (d, *J* = 2.0 Hz, 3 H).

2-(4-((2*S*)-4-((6-Amino-3-pyridinyl)sulfonyl)-2-(1-propyn-1-yl)-1-piperazinyl)phenyl)-1,1,1-trifluoro-4-pentyn-2-ol (64). A flame-dried, 50 mL, three-necked round-bottomed flask was charged with THF (8 mL) and ethynyltrimethylsilane (0.075 mL, 0.530 mmol). The reaction mixture was cooled to −78 °C, and *n*-butyllithium (2.5 M in hexanes, 0.220 mL, 0.530 mmol) was added at that temperature. The resulting mixture was stirred for 30 min at −78 °C. *tert*-Butyl (5-(((3*S*)-3-(1-propyn-1-yl)-4-(4-(2-(trifluoromethyl)-2-oxiranyl)phenyl)-1-piperazinyl)sulfonyl)-2-pyridinyl)-carbamate (**56**) (0.150 g, 0.265 mmol) in THF (5 mL) was added dropwise via an addition funnel. After the addition was complete, the reaction mixture was stirred at −78 °C for 10 min and then allowed to warm to room temperature and stirred at room temperature for an additional 20 h. The reaction was quenched with saturated aqueous NH_4Cl (3 mL). The reaction mixture was concentrated, and the residue was dissolved in MeOH (10 mL) and then treated with 2 M K_2CO_3 (3 mL) at room temperature for 2 h. The resulting mixture was partitioned between EtOAc (60 mL) and water (30 mL). The aqueous layer was extracted with EtOAc (50 mL). The combined organic layers were dried over $MgSO_4$, filtered, and concentrated. The crude product was purified by silica gel column chromatography (10−40% acetone in hexanes) to obtain *tert*-butyl (5-(((3*S*)-4-(4-(1-hydroxy-1-(trifluoromethyl)-3-butyn-1-yl)phenyl)-3-(1-propyn-1-yl)-1-piperazinyl)-sulfonyl)-2-pyridinyl)carbamate (**59**) (0.090 g, 57%) as an off-white solid. MS (ESI positive ion) *m*/*z*: calcd for $C_{28}H_{31}F_3N_4O_5S$, 592.197; found, 615.1 (M + Na). ^{1}H NMR (300 MHz, $CDCl_3$) δ 8.66 (d, *J* = 1.6 Hz, 1 H), 8.17−8.09 (m, 1 H), 8.08−7.96 (m, 1 H), 7.63 (s, 1 H), 7.46 (d, *J* = 8.8 Hz, 2 H), 6.95 (d, *J* = 9.1 Hz, 2 H), 4.43 (br s, 1 H), 3.79 (t, *J* = 11.1 Hz, 2 H), 3.48−3.31 (m, 2 H), 3.07 (dd, *J* = 2.3, 5.6 Hz, 2 H), 3.02−2.94 (m, 1 H), 2.87 (dd, *J* = 3.4, 11.1 Hz, 1 H), 2.78−2.62 (m, 1 H), 2.07 (t, *J* = 2.3 Hz, 1 H), 1.79 (d, *J* = 1.9 Hz, 3 H), 1.55 (s, 9 H).

A 20 mL vial was charged with compound **59** (0.025 g, 0.042 mmol), DCM (3.0 mL), and TFA (0.030 mL, 0.42 mmol). The vial was capped, and the mixture was stirred at room temperature for 2 h. The reaction mixture was partitioned between saturated aqueous $NaHCO_3$ (15 mL) and DCM (40 mL). The aqueous layer was extracted with DCM (2 × 30 mL), and the combined organic layers were dried over $MgSO_4$, filtered, and concentrated. The crude product was purified by silica gel column chromatography (0−5% MeOH in DCM) to afford 2-(4-((2*S*)-4-((6-amino-3-pyridinyl)sulfonyl)-2-(1-propyn-1-yl)-1-piperazinyl)phenyl)-1,1,1-trifluoro-4-pentyn-2-ol (**64**) (0.020 g, 96%) as an off-white solid. MS (ESI positive ion) *m*/*z*: calcd for $C_{23}H_{23}F_3N_4O_3S$, 492.114; found, 493.1 (M + 1). ^{1}H NMR (300 MHz, $CDCl_3$) δ 8.50 (d, *J* = 2.2 Hz, 1 H), 7.80 (dd, *J* = 2.3, 8.8 Hz, 1 H), 7.46 (d, *J* = 8.8 Hz, 2 H), 7.04−6.87 (m, 2 H), 6.56 (d, *J* = 8.8 Hz, 1 H), 5.15 (s, 2 H), 4.44 (br s, 1 H), 3.84−3.63 (m, 2 H), 3.47−3.33 (m, 2 H), 3.17−2.99 (m, 3 H), 2.85 (dd, *J* = 3.4, 11.2 Hz, 1 H), 2.79−2.62 (m, 1 H), 2.13−2.01 (m, 1 H), 1.79 (d, *J* = 1.9 Hz, 3 H).

2-(4-((2*S*)-4-((6-Amino-3-pyridinyl)sulfonyl)-2-(1-propyn-1-yl)-1-piperazinyl)phenyl)-1,1,1-trifluoro-4-hexyn-2-ol (65). A flame-dried, 50 mL, three-necked round-bottomed flask was charged with *tert*-butyl (5-(((3*S*)-4-(4-(1-hydroxy-1-(trifluoromethyl)-3-butyn-

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1-yl)phenyl)-3-(1-propyn-1-yl)-1-piperazinyl)sulfonyl)-2-pyridinyl)-carbamate (**59**) (0.060 g, 0.10 mmol) and THF (5.0 mL). The reaction mixture was cooled to 0 °C, and *n*-butyllithium (2.5 M in hexanes, 0.10 mL, 0.25 mmol) was added. The resulting mixture was stirred for 20 min, and then iodomethane (0.020 mL, 0.29 mmol) was added. The reaction mixture was allowed to warm to room temperature and stirred for an additional 2 h and then quenched with saturated aqueous NH_4Cl (3 mL), concentrated, and then partitioned between EtOAc (60 mL) and water (20 mL). The aqueous layer was extracted with EtOAc (2 × 30 mL), and the combined organic layers were dried over $MgSO_4$, filtered, and concentrated. The crude product was purified by silica gel column chromatography (0−40% EtOAc in hexanes) to afford *tert*-butyl (5-(((3*S*)-4-(4-(1-hydroxy-1-(trifluoromethyl)-3-pentyn-1-yl)phenyl)-3-(1-propyn-1-yl)-1-piperazinyl)sulfonyl)-2-pyridinyl)carbamate (**60**) (0.020 g, 33%) as an off-white solid. MS (ESI positive ion) *m*/*z*: calcd for $C_{29}H_{33}F_3N_4O_5S$, 606.212; found, 629.2 (M + Na). ^{1}H NMR (300 MHz, $CDCl_3$) δ 8.68 (d, *J* = 1.9 Hz, 1 H), 8.19−8.11 (m, 1 H), 8.08−7.99 (m, 1 H), 7.94 (s, 1 H), 7.45 (d, *J* = 8.8 Hz, 2 H), 6.93 (d, *J* = 8.9 Hz, 2 H), 4.43 (br s, 1 H), 3.78 (t, *J* = 10.9 Hz, 2 H), 3.47−3.32 (m, 2 H), 3.14−3.02 (m, 2 H), 3.01−2.83 (m, 2 H), 2.79−2.66 (m, 1 H), 1.78 (d, *J* = 1.9 Hz, 3 H), 1.75 (t, *J* = 2.3 Hz, 3 H), 1.56 (s, 9 H).

A 20 mL vial was charged with compound **60** (0.020 g, 0.03 mmol), DCM (2.0 mL), and TFA (0.030 mL, 0.33 mmol). The vial was capped and stirred at room temperature for 1 h. The reaction mixture was partitioned between saturated aqueous $NaHCO_3$ (15 mL) and DCM (40 mL). The aqueous layer was extracted with DCM (2 × 30 mL), and the combined organic layers were dried over $MgSO_4$, filtered, and concentrated. The crude product was purified by silica column chromatography (0−5% MeOH in DCM) to obtain 2-(4-((2*S*)-4-((6-amino-3-pyridinyl)sulfonyl)-2-(1-propyn-1-yl)-1-piperazinyl)phenyl)-1,1,1-trifluoro-4-hexyn-2-ol (**65**) (0.015 g, 90%) as an off-white solid. MS (ESI positive ion) *m*/*z*: calcd for $C_{24}H_{25}F_3N_4O_3S$, 506.160; found, 507.1 (M + 1). ^{1}H NMR (300 MHz, $CDCl_3$) δ 8.44 (s, 1 H), 7.87 (d, *J* = 7.2 Hz, 1 H), 7.46 (d, *J* = 8.6 Hz, 2 H), 6.95 (d, *J* = 8.8 Hz, 2 H), 6.76 (d, *J* = 8.9 Hz, 1 H), 6.11 (br s, 2 H), 4.45 (br s, 1 H), 3.76 (t, *J* = 10.3 Hz, 2 H), 3.46−3.33 (m, 2 H), 3.08 (dd, *J* = 2.5, 17.0 Hz, 2 H), 3.01−2.88 (m, 2 H), 2.78 (dd, *J* = 5.7, 10.4 Hz, 1 H), 1.79 (d, *J* = 1.8 Hz, 3 H), 1.75 (t, *J* = 2.3 Hz, 3 H).

(2*S*)-3-Amino-2-(4-((2*S*)-4-((6-amino-3-pyridinyl)sulfonyl)-2-(1-propyn-1-yl)-1-piperazinyl)phenyl)-1,1,1-trifluoro-2-propanol (67) and (2*R*)-3-Amino-2-(4-((2*S*)-4-((6-amino-3-pyridinyl)-sulfonyl)-2-(1-propyn-1-yl)-1-piperazinyl)phenyl)-1,1,1-trifluoro-2-propanol (68). A 20 mL vial was charged with *tert*-butyl(5-(((3*S*)-3-(1-propyn-1-yl)-4-(4-(2-(trifluoromethyl)-2-oxiranyl)-phenyl)-1-piperazinyl)sulfonyl)-2-pyridinyl)carbamate (**56**) (0.120 g, 0.212 mmol), DMF (3.0 mL), and ammonium hydroxide (0.165 mL, 1.23 mmol). The vial was sealed, and the reaction mixture was stirred at room temperature for 24 h. The reaction mixture was partitioned between EtOAc (60 mL) and water (30 mL), and the organic layer was washed with water (2 × 30 mL), dried over $MgSO_4$, filtered, and concentrated. To this crude intermediate were added DCM (10 mL) and TFA (0.160 mL, 2.12 mmol). The resulting mixture was stirred at room temperature for 2 h and then concentrated. The residue was partitioned between EtOAc (60 mL) and saturated $NaHCO_3$ (40 mL), and the aqueous layer was extracted with EtOAc (2 × 30 mL). The combined organic layers were dried over $MgSO_4$, filtered, and concentrated to afford 3-amino-2-(4-((2*S*)-4-((6-amino-3-pyridinyl)-sulfonyl)-2-(1-propyn-1-yl)-1-piperazinyl)phenyl)-1,1,1-trifluoro-2-propanol (**66**) (0.095 g) as a mixture of two diastereomers. The individual diastereomers were isolated using chiral SFC (Chiralpak AD-H Sepax column (150 mm × 21 mm, 5 μm)) using 45% (20 mM NH_3 in ethanol) in supercritical CO_2 (total flow was 75 mL/min). This produced the two compounds (**67** and **68**) with diastereomeric excess greater than 98%. The stereochemistry at carbinol was arbitrarily assigned.

(2*S*)-3-Amino-2-(4-((2*S*)-4-((6-amino-3-pyridinyl)sulfonyl)-2-(1-propyn-1-yl)-1-piperazinyl)phenyl)-1,1,1-trifluoro-2-propanol (**67**). MS (ESI positive ion) *m*/*z*: calcd for $C_{21}H_{24}F_3N_5O_3S$, 483.155; found, 484.1 (M + 1). ^{1}H NMR (300 MHz, $CDCl_3$) δ 8.49 (br s, 1 H), 7.76 (d, *J* = 7.5 Hz, 1 H), 7.44 (d, *J* = 7.6 Hz, 2 H), 6.92 (d, *J* = 8.3 Hz, 2 H), 6.51 (d, *J* = 8.8 Hz, 1 H), 4.93 (br s, 2 H), 4.39 (br s, 1 H), 3.72 (t, *J* = 9.3 Hz, 2 H), 3.60−3.42 (m, 1 H), 3.35 (br s, 2 H), 3.01 (br s, 1 H), 2.83 (d, *J* = 8.9 Hz, 1 H), 2.75−2.59 (m, *J* = 10.5 Hz, 1 H). Three exchangeable protons were not observed.

(2*R*)-3-Amino-2-(4-((2*S*)-4-((6-amino-3-pyridinyl)sulfonyl)-2-(1-propyn-1-yl)-1-piperazinyl)phenyl)-1,1,1-trifluoro-2-propanol (**68**). MS (ESI positive ion) *m*/*z*: calcd for $C_{21}H_{24}F_3N_5O_3S$, 483.155; found, 484.1 (M + 1). ^{1}H NMR (300 MHz, $CDCl_3$) δ 8.49 (br s, 1 H), 7.76 (d, *J* = 7.5 Hz, 1 H), 7.44 (d, *J* = 7.6 Hz, 2 H), 6.92 (d, *J* = 8.3 Hz, 2 H), 6.51 (d, *J* = 8.8 Hz, 1 H), 4.93 (br s, 2 H), 4.39 (br s, 1 H), 3.72 (t, *J* = 9.3 Hz, 2 H), 3.60−3.42 (m, 1 H), 3.35 (br s, 2 H), 3.01 (br s, 1 H), 2.83 (d, *J* = 8.9 Hz, 1 H), 2.75−2.59 (m, 1 H), 1.77 (s, 3 H). Three exchangeable protons were not observed.

2-(2-((2*S*)-4-((6-Amino-3-pyridinyl)sulfonyl)-2-(1-propyn-1-yl)-1-piperazinyl)-1,3-thiazol-5-yl)-1,1,1,3,3,3-hexafluoro-2-propanol (74). To a stirring suspension of 2-chloro-1,3-thiazole-5-carboxylic acid (**69**) (5.0 g, 31 mmol) in DCM (30 mL) at 0 °C under nitrogen was added oxalyl chloride (8.14 mL, 92 mmol) followed by DMF (0.118 mL, 1.53 mmol). The suspension was stirred for 3 h warming to 20 °C. The solvents were removed under reduced pressure and the residue was purified by silica gel chromatography (80 g), eluting with 0−20% EtOAc/hexanes to afford 2-chloro-1,3-thiazole-5-carbonyl chloride (**70**) (4.9 g, 88%) as a colorless oil. ^{1}H NMR (400 MHz, $CDCl_3$) δ 8.35 (s, 1 H). ^{13}C NMR (101 MHz, $CDCl_3$) δ 135.07, 150.76, 157.50, 161.45.

To a stirring solution of compound **70** (2.7 g, 15 mmol) and (trifluoromethyl)trimethylsilane (4.82 mL, 32.6 mmol) in 1,2-dimethoxyethane at −65 °C under argon was added tetramethylammonium fluoride (3.04 g, 32.6 mmol). The suspension had an exotherm to −52 °C and then stabilized. After the mixture was cooled to −60 °C, the cooling bath was removed. After the mixture was stirred for 20 h at room temperature, the reaction was quenched with 1 N HCl (50 mL) and the aqueous portion was extracted with EtOAc (50 mL). The separated organic layer was dried over $MgSO_4$, filtered, and concentrated under reduced pressure. The crude product was purified by silica gel chromatography (120 g), eluting with 0−30% EtOAc/Hex to afford 2-(2-chloro-1,3-thiazol-5-yl)-1,1,1,3,3,3-hexafluoro-2-propanol (2.23 g, 52.6%) as a white solid. MS (ESI positive ion) *m*/*z*: calcd for $C_6H_{2Cl}F_6NOS$, 284.945; found, 286.0 (M + 1).

To a stirring solution of 2-(2-chloro-1,3-thiazol-5-yl)-1,1,1,3,3,3-hexafluoro-2-propanol (2.23 g, 7.81 mmol) and DIPEA (3.00 mL, 17.2 mmol) in DCM (20 mL) at 0 °C under nitrogen was added (1,1-dimethylethyl)dimethylsilyl trifluoromethanesulfonate (2.15 mL, 9.37 mmol) over a 1 min period. After 30 min the reaction mixture was washed with 1 M KH_2PO_4 (20 mL). The organic phase was concentrated onto silica (15 g) under reduced pressure and then purified by silica gel chromatography (120 g), eluting with 0−20% EtOAc/Hex to afford 5-(1-((*tert*-butyl(dimethyl)silyl)oxy)-2,2,2-trifluoro-1-(trifluoromethyl)ethyl)-2-chloro-1,3-thiazole (**71**) (2.60 g, 83%) as a colorless oil. MS (ESI positive ion) *m*/*z*: calcd for $C_{12}H_{16Cl}F_6NOSSi$, 399.031; found, 400.0 (M + 1).

tert-Butyl (3*S*)-3-(1-propyn-1-yl)-1-piperazinecarboxylate[13] (**72**, 0.701 g, 3.13 mmol) and compound **71** (1.0 g, 2.5 mmol) were coupled according to the general amination method using RuPhos (0.058 g, 0.125 mmol), RuPhos first generation precatalyst (0.102 g, 0.125 mmol), and 1,4-dioxane to afford *tert*-butyl (3*S*)-4-(5-(1-((*tert*-butyl(dimethyl)silyl)oxy)-2,2,2-trifluoro-1-(trifluoromethyl)ethyl)-1,3-thiazol-2-yl)-3-(1-propyn-1-yl)-1-piperazinecarboxylate (**73**) (0.64 g, 44%) as a colorless oil. MS (ESI positive ion) *m*/*z*: calcd for $C_{24}H_{35}F_6N_3O_3SSi$, 587.207; found, 588.0 (M + 1). ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.12 (s, 1 H), 4.64 (br s, 1 H), 3.94−4.18 (m, 2 H), 3.38 (br s, 1 H), 3.24−3.36 (m, 1 H), 3.03 (br s, 1 H), 2.82 (br s, 1 H), 1.64 (d, *J* = 1.76 Hz, 3 H), 1.35 (s, 9 H), 0.81 (s, 9 H), 0.00 (s, 6 H).

Compound **73** (0.64 g, 1.1 mmol) was deprotected according to the general Boc-deprotection method. The crude product was treated with *tert*-butyl (5-(chlorosulfonyl)-2-pyridinyl)carbamate (**9**) (0.398 g, 1.36 mmol) according to the general sulfonamide formation method using

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DIPEA (1.9 mL, 11 mmol) to afford *tert*-butyl (5-(((3*S*)-4-(5-(1-((*tert*-butyl(dimethyl)silyl)oxy)-2,2,2-trifluoro-1-(trifluoromethyl)-ethyl)-1,3-thiazol-2-yl)-3-(1-propyn-1-yl)-1-piperazinyl)sulfonyl)-2-pyridinyl)carbamate (0.40 g, 49%) as a colorless oil. MS (ESI positive ion) *m*/*z*: calcd for $C_{29}H_{39}F_6N_5O_5S_2Si$, 743.207; found, 744.0 (M + 1).

To a stirring solution of *tert*-butyl (5-(((3*S*)-4-(5-(1-((*tert*-butyl-(dimethyl)silyl)oxy)-2,2,2-trifluoro-1-(trifluoromethyl)ethyl)-1,3-thiazol-2-yl)-3-(1-propyn-1-yl)-1-piperazinyl)sulfonyl)-2-pyridinyl)-carbamate (0.400 g, 0.538 mmol) in MeCN (4 mL) at 20 °C under argon was added HF−pyridine complex (70% HF, 301 μL, 2.42 mmol). The solution was stirred overnight at 20 °C. To the mixture was added TFA (5 mL), and the mixture was stirred for 24 h at 20 °C. The solvents were removed under reduced pressure, and the residue was partitioned between 9:1 $CHCl_3$/IPA (30 mL) and 1 M KH_2PO_4 (pH 5). The separated organic layer was then dried over $MgSO_4$, filtered, and concentrated under reduced pressure. The crude material was purified by silica gel chromatography (12 g), eluting with 0−8% 2 M NH_3 in MeOH/DCM to afford 2-(2-((2*S*)-4-((6-amino-3-pyridinyl)sulfonyl)-2-(1-propyn-1-yl)-1-piperazinyl)-1,3-thiazol-5-yl)-1,1,1,3,3,3-hexafluoro-2-propanol (**74**) (0.220 g, 77%) as a white foam. MS (ESI positive ion) *m*/*z*: calcd for $C_{17}H_{18}F_6N_5O_3S_2$, 529.068; found, 530.0 (M + 1). ^{1}H NMR (400 MHz, DMSO-d_6) δ 9.12 (s, 1 H), 8.23 (d, *J* = 2.4 Hz, 1 H), 7.63 (dd, *J* = 9.0, 2.5 Hz, 1 H), 7.38 (s, 1 H), 7.05 (br s, 2 H), 6.54 (d, *J* = 8.8 Hz, 1 H), 4.99 (br s, 1 H), 3.75 (d, *J* = 12.3 Hz, 1 H), 3.56−3.69 (m, 2 H), 3.26−3.37 (m, 1 H), 2.59 (dd, *J* = 11.6, 3.4 Hz, 1 H), 2.37−2.49 (m, 1 H), 1.80 (d, *J* = 2.2 Hz, 3 H).

2-(2-(4-((6-Amino-3-pyridinyl)sulfonyl)-2-(1-propyn-1-yl)-1-piperazinyl)-1,3-thiazol-5-yl)-1,1,1-trifluoro-2-propanol (80). To a stirring suspension of 2-chloro-1,3-thiazole-5-carboxylic acid (**69**) (5.0 g, 31 mmol) in DCM (30 mL) at 0 °C under nitrogen were added oxalyl chloride (8.1 mL, 92 mmol) and DMF (0.118 mL, 1.53 mmol). The suspension was stirred for 5 h warming to 20 °C to create a solution. The solvents were then removed under reduced pressure and redissolved in DCM (30 mL). To the solution was added DIPEA (16 mL, 92 mmol), and then the mixture was cooled to 0 °C under nitrogen. To the reaction mixture *N*,*O*-dimethylhydroxylamine hydrochloride (2.98 g, 30.6 mmol) was added at once. After 15 min the reaction mixture was diluted with 1 M KH_2PO_4 (75 mL). The organic phase was dried over $MgSO_4$, filtered, and concentrated under reduced pressure. The crude material was purified by silica gel chromatography (120 g), eluting with 20−50% EtOAc/Hex to afford 2-chloro-*N*-methoxy-*N*-methyl-1,3-thiazole-5-carboxamide (**75**) (5.31 g, 84%) as a yellow solid. MS (ESI positive ion) *m*/*z*: calcd for $C_6H_7ClN_2O_2S$, 205.992; found, 206.9 (M + 1).

A suspension of benzyl 3-(1-propyn-1-yl)-1-piperazinecarboxylate (**6**)[13] (1.0 g, 3.9 mmol), compound **75** (1.20 g, 5.81 mmol), and DIPEA (1.01 mL, 5.81 mmol) in DMF (5 mL) was heated to 100 °C with stirring for 20 h. The reaction mixture was allowed to cool to room temperature and partitioned between EtOAc (50 mL) and 1 M KH_2PO_4 (80 mL). The organic phase was then dried over $MgSO_4$, filtered, concentrated under reduced pressure, and then purified by silica gel chromatography (80 g), eluting with 0−30% acetonitrile in DCM to afford benzyl 4-(5-(methoxy(methyl)carbamoyl)-1,3-thiazol-2-yl)-3-(1-propyn-1-yl)-1-piperazinecarboxylate (**76**) (0.58 g, 35%) as a colorless oil. MS (ESI positive ion) *m*/*z*: calcd for $C_{21}H_{24}N_4O_4S$, 428.152; found, 429.0 (M + 1). ^{1}H NMR (400 MHz, $CDCl_3$) δ 8.06 (s, 1 H), 7.28−7.43 (m, 5 H), 5.08−5.29 (m, 2 H), 4.95 (br s, 1 H), 4.17−4.43 (m, 2 H), 3.75 (s, 3 H), 3.61−3.72 (m, 1 H), 3.44−3.58 (m, 1 H), 3.32 (s, 3 H), 3.01−3.29 (m, 2 H), 1.71 (br s, 3 H).

To a solution of compound **76** (0.550 g, 1.28 mmol) in THF (5 mL) at 0 °C under nitrogen was added methylmagnesium bromide (3.0 M in diethyl ether, 1.28 mL, 3.85 mmol) dropwise. The reaction mixture was stirred for 30 min, then slowly quenched with saturated NH_4Cl (20 mL) and EtOAc (30 mL). The organic phase was dried over $MgSO_4$, filtered, concentrated under reduced pressure, and then purified by silica gel chromatography (40 g), eluting with 0−40% EtOAc in hexanes to afford benzyl 4-(5-acetyl-1,3-thiazol-2-yl)-3-(1-propyn-1-yl)-1-piperazinecarboxylate (0.392 g, 80%) as a colorless oil. MS (ESI positive ion) *m*/*z*: calcd for $C_{20}H_{21}N_3O_3S$, 383.130; found, 384.0 (M + 1). ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.83 (s, 1 H), 7.29−7.42 (m, 5 H), 5.10−5.29 (m, 2 H), 4.92−5.02 (m, 1 H), 4.24−4.41 (m, 2 H), 3.64−3.72 (m, 1 H), 3.49−3.62 (m, 1 H), 2.96−3.30 (m, 2 H), 2.45 (s, 3 H), 1.70 (br s, 3 H).

To a stirring solution of benzyl 4-(5-acetyl-1,3-thiazol-2-yl)-3-(1-propyn-1-yl)-1-piperazinecarboxylate (0.380 g, 0.991 mmol) and trifluoromethyltrimethylsilane (220 μL, 1.49 mmol) in THF (3 mL) under nitrogen at 0 °C was added tetramethylammonium fluoride (0.111 g, 1.19 mmol) at once. After the mixture was stirred for 30 min at 0 °C, the reaction was quenched with 10% citric acid (20 mL) and the aqueous portion was extracted with EtOAc (30 mL). The organic phase was dried over $MgSO_4$, filtered, concentrated onto silica (15 g), and then purified by silica gel chromatography (40 g), eluting with 20−60% EtOAc in hexanes to afford benzyl 3-(1-propyn-1-yl)-4-(5-(2,2,2-trifluoro-1-hydroxy-1-methylethyl)-1,3-thiazol-2-yl)-1-piperazinecarboxylate (**77**) (0.068 g, 15%) as a colorless oil. MS (ESI positive ion) *m*/*z*: calcd for $C_{21}H_{22}F_3N_3O_3S$, 453.133; found, 454.1 (M + 1).

Compound **77** (0.070 g, 0.154 mmol) was deprotected according to the general Cbz-deprotection method A. The crude product was treated with 6-chloro-3-pyridinesulfonyl chloride**78**[19] (0.0491 g, 0.232 mmol) according to the general sulfonamide formation method using DIPEA (0.27 mL, 1.5 mmol) to afford 2-(2-(4-((6-chloro-3-pyridinyl)sulfonyl)-2-(1-propyn-1-yl)-1-piperazinyl)-1,3-thiazol-5-yl)-1,1,1-trifluoro-2-propanol (**79**) (0.058 g, 76%) as a colorless film. MS (ESI positive ion) *m*/*z*: calcd for $C_{18}H_{18}ClF_3N_4O_3S_2$, 494.046; found, 494.9 (M + 1).

A solution of compound **79** (0.058 g, 0.117 mmol) in EtOH (1.5 mL) and NH_4OH (28% in water, 1 mL) was heated to 120 °C with microwave apparatus for 3 h. The reaction mixture was allowed to cool to room temperature and partitioned between 9:1 $CHCl_3$/IPA (15 mL) and 5% $NaHCO_3$ (7 mL). The aqueous layer was further extracted with 9:1 $CHCl_3$/IPA (10 mL). The combined organics were dried over $MgSO_4$, filtered, concentrated under reduced pressure, and then purified by silica gel chromatography (4 g), eluting with 0−6% (2 M NH_3 in MeOH) in DCM to afford 2-(2-(4-((6-amino-3-pyridinyl)sulfonyl)-2-(1-propyn-1-yl)-1-piperazinyl)-1,3-thiazol-5-yl)-1,1,1-trifluoro-2-propanol (**80**) (0.013 g, 23%) as a colorless film. MS (ESI positive ion) *m*/*z*: calcd for $C_{18}H_{20}F_3N_5O_3S_2$, 475.096; found, 476.0 (M + 1). ^{1}H NMR (400 MHz, DMSO-d_6) δ 8.22 (d, *J* = 2.5 Hz, 1 H), 7.60 (s, 1 H), 7.21 (d, *J* = 3.9 Hz, 1 H), 7.01 (s, 2 H), 6.97 (d, *J* = 1.2 Hz, 1 H), 6.52 (d, *J* = 8.8 Hz, 1 H), 4.87−4.99 (m, 1 H), 3.62 (br s, 3 H), 3.31 (m, 1 H), 2.52−2.60 (m, 1 H), 2.31−2.46 (m, 1 H), 1.79 (t, *J* = 2.4 Hz, 3 H), 1.66 (s, 3 H). HPLC purity at 254 nm, 89.0%

2-(2-((2*S*)-4-((6-Amino-3-pyridinyl)sulfonyl)-2-(1-propyn-1-yl)-1-piperazinyl)-5-pyrimidinyl)-1,1,1,3,3,3-hexafluoro-2-propanol (88). A 1 L round-bottomed flask was charged with 2-piperazinone (**81**) (10.2 g, 102 mmol) and DCM (250 mL). 6-Chloro-3-pyridinesulfonyl chloride (**78**)[19] (21.8 g, 103 mmol) was added in portions at 0 °C under argon stream. Then Et_3N (32.0 mL, 230 mmol) in DCM (50 mL) was added via an additional funnel over 30 min at 0 °C. The ice−water bath was removed, and the milky mixture was stirred at room temperature for 30 min. The mixture was concentrated, and the residue was taken into saturated aqueous $NaHCO_3$ (200 mL). The mixture was stirred at room temperature for 30 min. The white precipitate was collected via filtration. The filter cake was washed with water (3 × 200 mL), air-dried, and then dried in a vacuum oven (60 °C, 50 mmHg) for 18 h to afford 4-((6-chloro-3-pyridinyl)sulfonyl)-2-piperazinone (**82**) (27.5 g, 97%) as an off-white solid. MS (ESI positive ion) *m*/*z*: calcd for $C_9H_{10}ClN_3O_3S$, 275.013; found, 276.0 (M + 1). ^{1}H NMR (400 MHz, DMSO-d_6) δ 8.81 (d, *J* = 2.2 Hz, 1 H), 8.24 (dd, *J* = 2.5, 8.4 Hz, 1 H), 8.05 (br s, 1 H), 7.79 (d, *J* = 8.4 Hz, 1 H), 3.59 (s, 2 H), 3.31−3.25 (m, 2 H), 3.21−3.15 (m, 2 H).

A 1 L round-bottomed flask was charged with compound **82** (27.5 g, 100 mmol), DMAP (1.49 g, 12.2 mmol), TEA (30.0 mL, 215 mmol), and DCM (300 mL). Di-*tert*-butyl dicarbonate (44.01 g, 202 mmol) was added in portions at room temperature. The mixture was stirred at room temperature for 1.5 h. Additional DMAP (1.57 g, 12.8 mmol) was added, and stirring was resumed for 1.5 h (3 h total). The reaction mixture was concentrated, and the residue was taken into

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EtOAc (500 mL). The slurry was washed with 1 N HCl (2 × 200 mL), water (200 mL), and saturated aqueous NaCl (200 mL). The EtOAc phase was filtered to collect a white solid. The solid was washed with EtOAc (2 × 100 mL). The filtrate and washes were combined and concentrated. EtOAc (100 mL) was added to the residue to induce the second crop of the product. The resulting white precipitate was collected via filtration, and the filter cake was washed with EtOAc (2 × 50 mL). The two crops were combined to give *tert*-butyl 4-((6-chloro-3-pyridinyl)sulfonyl)-2-oxo-1-piperazinecarboxylate (**83**) (32.0 g, 85%) as a white solid. MS (ESI positive ion) *m*/*z*: calcd for $C_{14}H_{18}ClN_3O_5S$, 375.066; found, 398.0 (M + Na). ^{1}H NMR (400 MHz, $CDCl_3$) δ 8.81 (d, *J* = 2.0 Hz, 1 H), 8.03 (dd, *J* = 2.3, 8.4 Hz, 1 H), 7.55 (d, *J* = 8.4 Hz, 1 H), 3.94−3.80 (m, 4 H), 3.44 (t, *J* = 5.5 Hz, 2 H), 1.52 (s, 9 H).

A 1 L round-bottomed flask was charged with compound **83** (11.05 g, 29.4 mmol) and 100 mL of THF. After the mixture was cooled to 0 °C, bromo(1-propyn-1-yl)magnesium (0.5 M in THF, 88 mL, 44.1 mmol) was added. Soon after the finish of the addition, the reaction was quenched with saturated aqueous NH_4Cl and the aqueous portion was extracted with EtOAc. The combined extracts were dried over $MgSO_4$, filtered, and concentrated to give an oil. To this was added DCM (30 mL). After the mixture was cooled to 0 °C, TFA (33.5 g, 294 mmol) was added. After 15 min, sodium triacetoxyhydroborate (24.9 g, 118 mmol) was added portionwise at 0 °C. The mixture was stirred at room temperature for 1.5 h, and then the mixture was concentrated. To this oil was added DCM (100 mL), and then the mixture was neutralized with 5 N NaOH to pH 6−7. The layers were separated, and the organics were dried over $MgSO_4$, filtered, and concentrated. The resulting oil was purified by silica gel column chromatography (10−100% EtOAc in hexanes) to give 1-((6-chloro-3-pyridinyl)sulfonyl)-3-(1-propyn-1-yl)piperazine (**85**) (7.13 g, 23.8 mmol, 81%) as a brown brittle foam. MS (ESI positive ion) *m*/*z*: calcd for $C_{12}H_{14}ClN_3O_2S$, 299.050; found, 300.1(M + 1).

A 350 mL pressure tube was charged with compound **85** (7.13 g, 23.8 mmol), 2-(2-chloro-5-pyrimidinyl)-1,1,1,3,3,3-hexafluoro-2-propanol (**86**),[13] (7.34 g, 26.2 mmol), NMP (30 mL), and DIPEA (8.27 mL, 47.6 mmol). The tube was sealed and heated at 130 °C for 8 h. The mixture was allowed to cool to room temperature and then diluted with water (100 mL) and EtOAc (200 mL). The biphasic mixture was transferred to a separatory funnel, and the aqueous layer was removed. The remaining organics were washed with water (2 × 150 mL), saturated aqueous NaCl (150 mL), dried over $MgSO_4$, filtered, and concentrated to give a brown oil. Purification via silica gel column chromatography (0−50% EtOAc in hexanes) gave crude 2-(2-(4-((6-chloro-3-pyridinyl)sulfonyl)-2-(1-propyn-1-yl)-1-piperazinyl)-5-pyrimidinyl)-1,1,1,3,3,3-hexafluoro-2-propanol (~7.5 g) as an off-white brittle foam. To this was added concentrated NH_4OH (20 mL) and EtOH (30 mL). The mixture was heated in a sealed tube at 135 °C for 8 h. The mixture was allowed to cool to room temperature and then concentrated. To this solution was added MeOH (40 mL) and sodium borohydride (0.900 g, 23.8 mmol) to reduce the methyl ketone byproduct formed via hydration of the triple bond, thereby facilitating the removal of this byproduct in the purification process. After 5 min, the reaction mixture was concentrated. The residue was dissolved in EtOAc and washed with water. The organics were separated, dried over $MgSO_4$, filtered, and concentrated. The resulting oil was purified by silica gel column chromatography (0−50% EtOAc in hexanes) to give 2-(2-(4-((6-amino-3-pyridinyl)sulfonyl)-2-(1-propyn-1-yl)-1-piperazinyl)-5-pyrimidinyl)-1,1,1,3,3,3-hexafluoro-2-propanol (**87**) (4.15 g, 33%) as an off-white foam. MS (ESI positive ion) *m*/*z*: calcd for $C_{19}H_{18}F_6N_6O_3S$, 524.107; found, 525.1 (M + 1).

The racemate **87** (0.082 g, 0.16 mmol) was separated using chiral SFC as follows: AD-H (5 μm, 21 mm × 250 mm) column, using 70% carbon dioxide−30% MeOH; flow rate was 70 mL/min. This procedure produced 2-(2-((2*S*)-4-((6-amino-3-pyridinyl)sulfonyl)-2-(1-propyn-1-yl)-1-piperazinyl)-5-pyrimidinyl)-1,1,1,3,3,3-hexafluoro-2-propanol (**88**). The absolute stereochemistry at the piperazine carbon next to the N−Ar was assigned to be (*S*) configuration based on the result obtained in the biochemical assay. MS (ESI positive ion) *m*/*z*: calcd for $C_{19}H_{18}F_6N_6O_3S$, 524.107; found, 525.0 (M + 1). ^{1}H NMR (400 MHz, $CDCl_3$) δ 8.59 (s, 2 H), 8.43 (d, *J* = 2.15 Hz, 1 H), 7.77 (dd, *J* = 2.25, 8.71 Hz, 1 H), 6.54 (d, *J* = 8.80 Hz, 1 H), 5.75 (br s, 1 H), 5.05−5.18 (m, 2 H), 4.69 (d, *J* = 13.30 Hz, 1 H), 3.75−3.91 (m, 2 H), 3.43−3.58 (m, 1 H), 2.63 (dd, *J* = 3.72, 11.15 Hz, 1 H), 2.49 (dt, *J* = 3.03, 11.69 Hz, 1 H), 1.82 (d, *J* = 2.15 Hz, 3 H). One exchangeable proton was not observed.

■ ASSOCIATED CONTENT

Ⓢ Supporting Information

Standard deviations for the in vitro data listed in Tables 1−3; VCD data for compounds **89** and **90**; and synthetic procedures for the preparation of piperazine intermediates **4**, **5**, **6**, and **72**, sulfonyl chloride **9**, and aryl halides intermediates that were not commercially available. This material is available free of charge via the Internet at http://pubs.acs.org.

Accession Codes

The cocrystal structures of hGKRP + compounds **51**, **65**, **15**, and **24** have been deposited in the Protein Data Bank with PDB codes 4OHM, 4OHK, 4OHP, and 4OHO, respectively.

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Notes

The authors declare no competing financial interest.

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■ ABBREVIATIONS USED

Ac, acetyl; BINAP, 2,2′-bis(diphenylphosphino)-1,1′-binaphthyl; Bn, benzyl; Boc, *tert*-butyloxycarbonyl; Cbz, carboxybenzyl; CL, clearance; DavePhos, 2′-(biphenylphosphino)- *N*,*N*-dimethyl-[1,1′-biphenyl]-2-amine; dba, 1,5-diphenylpenta-1,4-dien-3-one; DCM, dichloromethane; DIPEA, diisopropylethylamine; DMAP, 4-dimethylaminopyridine; DME, 1,2-dimethoxyethane; DMF, *N*,*N*-dimethylformamide; DMSO, dimethyl sulfoxide; Et_3N, triethylamine; EtOAc, ethyl acetate; EtOH, ethyl alcohol; GK, glucokinase; GKA, glucokinase activator; GKRP, glucokinase regulatory protein; IPA, isopropyl alcohol; iv, intravenous; JohnPhos, 2-biphenylyl(di-*tert*-butyl)-phosphane; MeOH, methyl alcohol; NMP, 1-methyl-2-pyrrolidinone; PD, pharmacodynamic; po, orally; RLM, rat liver microsome; RuPhos, 2-dicyclohexyl(2′,6′-diisopropoxybiphenyl-2-yl)phosphine; RuPhos first generation precatalyst, chloro(2-dicyclohexylphosphino-2′,6′-diisopropoxy-1,1′-biphenyl)[2-(2-aminoethylphenyl)]palladium(II); SFC, supercritical fluid chromatography; TBAF, tetrabutylammonium fluoride; TBS, *tert*-butyldimethylsilyl; Tf, trifluoromethanesulfonyl; TFA, trifluoroacetic acid; THF, tetrahydrofuran; Ts, *p*-toluenesulfonyl; X-Phos, 2-dicyclohexylphosphino-2′,4′,6′-triisopropyl-1,1′-biphenyl

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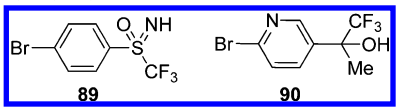

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(21) CL_{int} (μL min^{-1} mg^{-1}) is estimated from parent compound (1 μM) remaining following a 30 min incubation in rat liver microsomes in potassium phosphate buffered (66.7 mM) with NADPH (1 mM) at 37 °C for 30 min. Under these conditions, a cutoff of <100 μL min^{-1} (mg protein)$^{-1}$ was considered desirable.
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