3-Hydroxymethyl-7-(*N*-substituted aminosulfonyl)-1,2,3,4-tetrahydroisoquinoline Inhibitors of Phenylethanolamine *N*-Methyltransferase that Display Remarkable Potency and Selectivity[1]

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Six 3-hydroxymethyl-7-(*N*-substituted aminosulfonyl)-1,2,3,4-tetrahydroisoquinolines (**16**−**21**) were synthesized and evaluated for their phenylethanolamine *N*-methyltransferase (PNMT) inhibitory potency and affinity for the α_2-adrenoceptor. The addition of nonpolar substituents to the sulfonamide nitrogen of **9** (3-CH_2OH-7-SO_2NH_2-THIQ) led to inhibitors (**16**−**21**) that have high PNMT inhibitory potency and high selectivity, and most of these (**16**−**21**) are predicted, on the basis of their calculated log *P* values, to be able to penetrate the blood−brain barrier. Compounds *N*-trifluoroethyl sulfonamide **20** (PNMT K_i = 23 nM) and *N*-trifluoropropyl sulfonamide **21** (PNMT K_i = 28 nM) are twice as potent at inhibiting PNMT compared to **9** and display excellent selectivity (α_2 K_i/PNMT $K_i \geq$ 15 000).

Introduction

Epinephrine (Epi, **2**) was identified within the central nervous system (CNS) 50 years ago.[2] Since that time, it has been implicated in some of the neurodegeneration seen in Alzheimer's disease,[3,4] as well as in the regulation of blood pressure,[5] respiration,[6,7] body temperature,[6,7] the α_1-adrenoceptor,[8] and the α_2-adrenoceptor.[9,10] However, its role in the CNS still remains unclear. One approach to study the effects of CNS Epi (**2**) would be to inhibit the enzyme phenylethanolamine *N*-methyltransferase (PNMT; EC 2.1.1.28).[11] This enzyme catalyzes the last step in the biosynthetic pathway of Epi (**2**) in which an activated methyl group from *S*-adenosyl-L-methionine (**3**) is transferred to the primary amine of NE (**1**) to produce Epi (**2**) and *S*-adenosyl-L-homocysteine (AdoHcy, **4**; Figure 1).

Some of the first inhibitors of PNMT discovered were benzylamines.[12] Constraining benzylamines to 1,2,3,4-tetrahydroisoquinolines (THIQs) increases their PNMT inhibitory potency.[13] SK&F 29661[14] (**5**) and SK&F 64139[15] (**6**) were some of the first examples of potent THIQ-type PNMT inhibitors (Table 1). However, use of these compounds as physiological probes to elucidate the role of CNS Epi (**2**) has been complicated for two different reasons. First, SK&F 29661 (**5**) is quite polar (calculated log $P = -0.29$) and does not cross the blood−brain barrier (BBB).[14] A previous study from our lab using a BBB model[16,17] on a small set of THIQs[18] gave a good ($r = 0.79$) correlation between the calculated log *P*[19,20] of the compound and its permeability coefficient. This correlation indicated that THIQs with a calculated log *P* value of approximately 0.5 or greater should be able to penetrate into the CNS. The second concern is that SK&F 64139 (**6**), as well as many other THIQ-type PNMT inhibitors, shows affinity for the α_2-adrenoceptor, which complicates interpretation of the biochemical

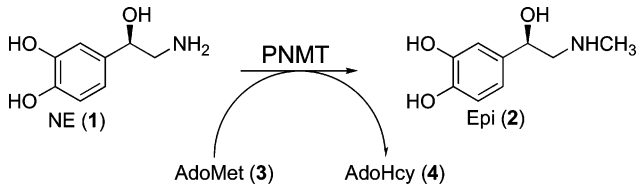

Figure 1. The terminal step in Epi (**2**) biosynthesis is the transfer of a methyl group from AdoMet (**3**) to NE (**1**) to form Epi (**2**) and the cofactor product AdoHcy (**4**).

results. Previous studies have shown that 3,7-disubstitution on the THIQ nucleus increases selectivity for PNMT versus the α_2-adrenoceptor.[21] For example, the selectivity of **8** is much greater than that of either monosubstituted **5** or **7**.[18] As such, the objective of this research is to develop a 3,7-disubstituted-THIQ-type PNMT inhibitor that is potent, selective, and capable of penetrating the BBB.

(±)-7-Aminosulfonyl-3-fluoromethyl-THIQ (**8**) is a highly potent and selective PNMT inhibitor, but this compound is predicted to be too polar (calculated log $P = 0.00$) to cross the BBB.[18] An approach to increase the lipophilicity of **8** that proved to be highly successful was the addition of nonpolar substituents to the sulfonamide nitrogen of **8**.[22] Not only are these inhibitors predicted to penetrate the BBB, but *N*-trifluoroethyl sulfonamide **14** and *N*-trifluoropropyl sulfonamide **15** retained PNMT inhibitory potency and dramatically increased selectivity compared to parent **8**. Since the addition of nonpolar substituents to **8** was successful, it was of interest to determine if this same approach could be applied to other PNMT inhibitors that are highly potent and selective but incapable of penetrating the BBB. In this paper, this strategy was applied to (±)-7-aminosulfonyl-3-hydroxymethyl-THIQ (**9**; PNMT K_i = 52 nM, α_2 K_i/PNMT K_i = 27 000) to increase its lipophilicity with the expectation that *N*-substituted analogues of **9** could be highly potent and selective PNMT inhibitors. Of the 27 (±)-3-fluoromethyl-7-(*N*-substituted aminosulfonyl)-THIQs prepared in the earlier study, six (**10**−**15**)

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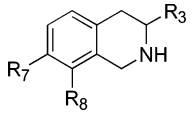
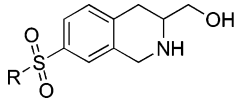
Table 1. In Vitro PNMT and α_2-Adrenoceptor Affinities of Some PNMT Inhibitors



compd	R_3	R_7	R_8	K_i (μM) ± SEM hPNMT	K_i (μM) ± SEM α_2[a]	selectivity (α_2/hPNMT)	ClogP[b]
5[c,d]	H	SO_2NH_2	H	0.28 ± 0.02	100 ± 10	360	−0.29
6[d,e]	H	Cl	Cl	0.0031 ± 0.0006	0.021 ± 0.005	7	2.75
7[d,f]	CH_2F	H	H	0.82 ± 0.04	3.8 ± 0.1	4.6	1.84
8[d,f]	CH_2F	SO_2NH_2	H	0.15 ± 0.01	680 ± 10	4500	0.00
9[d,g]	CH_2OH	SO_2NH_2	H	0.052 ± 0.004	1400 ± 200	27000	−0.93
10[h]	CH_2F	$SO_2NHCH_2CH_3$	H	1.4 ± 0.1	550 ± 60	390	1.15
11[h]	CH_2F	$SO_2NH(CH_2)_2CH_3$	H	1.7 ± 0.2	610 ± 60	360	1.67
12[h]	CH_2F	$SO_2N(CH_2CH_2)_2S$	H	1.2 ± 0.2	190 ± 20	160	1.71
13[d,f]	CH_2F	SO_2NH-Ph-4-Cl	H	0.27 ± 0.02	140 ± 20	520	3.18
14[h]	CH_2F	SO_2NHCH_2CF3	H	0.13 ± 0.02	1200 ± 100	9200	1.41[i]
15[h]	CH_2F	$SO_2NH(CH_2)_2CF_3$	H	0.22 ± 0.02	660 ± 80	3000	1.39[i]

[a] In vitro activities reported for the inhibition of binding of [^{3}H]clonidine at the α_2-adrenoceptor. [b] Calculated log *P*; refs 19 and 20. [c] Reference 14. [d] Biochemical data has been reported earlier for bovine PNMT. Here we report data for recombinant human PNMT. [e] Reference 15. [f] Reference 18. [g] Reference 21. [h] Reference 22. [i] The trend in ClogP values is consistent with positioning a trifluoromethyl group next to a heteroatom; ref 33.

Table 2. In Vitro Activities of (±)-3-Hydroxymethyl-7-(*N*-substituted aminosulfonyl)-1,2,3,4-tetrahydroisoquinolines



compd	R	K_i (μM) ± SEM hPNMT	K_i (μM) ± SEM α_2[a]	selectivity (α_2/hPNMT)	ClogP[b]
16	$NHCH_2CH_3$	0.17 ± 0.01	210 ± 20	1200	0.22
17	$NHCH_2CH_2CH_3$	0.19 ± 0.01	230 ± 20	1200	0.75
18	$N(CH_2CH_2)_2S$	0.23 ± 0.01	320 ± 30	1400	0.79
19	NH(4-Cl-Ph)	0.063 ± 0.002	53 ± 5	840	2.26
20	$NHCH_2CF_3$	0.023 ± 0.002	340 ± 40	15000	0.48[c]
21	$NHCH_2CH_2CF_3$	0.028 ± 0.001	520 ± 50	19000	0.46[c]

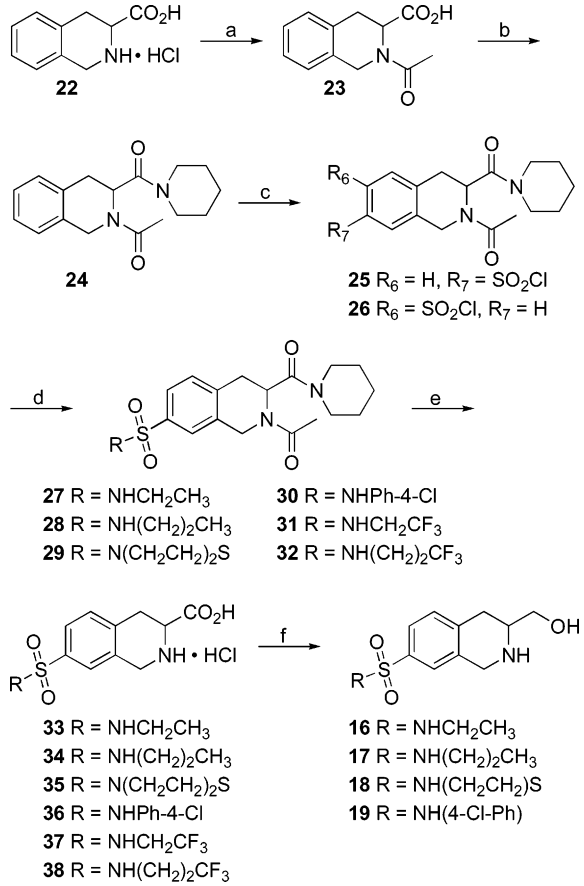
[a] In vitro activities reported for the inhibition of binding of [^{3}H]clonidine at the α_2-adrenoceptor. [b] Calculated log *P*. [c] The trend in ClogP values is consistent with positioning a trifluoromethyl group next to a heteroatom; ref 33.

displayed high PNMT inhibitory potency and selectivity (Table 1). Here we report the synthesis and biochemical evaluation of the *N*-substituted analogues of **9** (**16**−**21**) with these same substituents (Table 2).

Chemistry

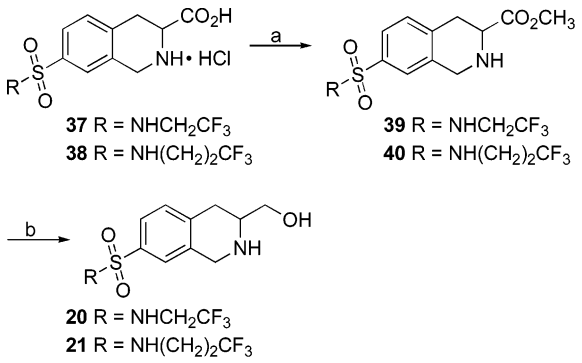
The route for the synthesis of **16**−**21** is shown in Schemes 1 and 2. Amino acid **22** was protected as the acetamide (**23**) by treatment with acetyl chloride in an acetone/NaOH mixture.[23] The acid of **23** was protected as the piperidinamide (**24**). Treatment of **24** with chlorosulfonic acid afforded two sulfonyl chloride regioisomers, **25** (44%) and **26** (15%), that were easily separated by flash chromatography. Two-dimensional NMR studies (HMQC, HMBC) indicated that **25** and **26** were substituted at the 7- and 6-positions of the aromatic ring, respectively. Compound **25** was carried forward in the synthesis and the substitution pattern of **25** was further confirmed from an X-ray crystal structure of THIQ **21**. Compound **25** was treated with the requisite amines to form sulfonamides **27**−**32**. Simultaneous hydrolysis of the acetamide and piperidinamide of **27**−**32** was accomplished with 6 N HCl to produce **33**−**38**. The crude amino acid hydrochloride salts (**33**−**36**) were directly reduced with BH_3·THF to form the corresponding THIQs (**16**−**19**). Compounds **37** and **38** were insoluble in the solvent (THF) used for the reduction and were converted to their methyl esters (**39** and **40**) with thionyl chloride and methanol, followed by reduction with $LiBH_4$ to produce the corresponding THIQs (**20** and **21**).

Scheme 1[a]



[a] Reagents: (a) acetyl chloride, 2 N NaOH; (b) BOP, HOBt, piperidine; (c) chlorosulfonic acid; (d) 1° or 2° amine; (e) 6 N HCl; (f) BH_3·THF.

Scheme 2[a]



[a] Reagents: (a) $SOCl_2$, MeOH; (b) $LiBH_4$.

Biochemistry

In the current study, human PNMT (hPNMT) with a C-terminal hexahistidine tag was expressed in *Escherichia coli*.[22,24] The radiochemical assay conditions, previously reported for the bovine enzyme,[25] were modified to account for the high binding affinity of some inhibitors. Inhibition constants were determined using four concentrations of phenylethanolamine as the variable substrate and three concentrations of inhibitor.

α_2-Adrenergic receptor binding assays were performed using cortex obtained from male Sprague–Dawley rats.[26] [^{3}H]Clonidine was used as the radioligand to define the specific binding and phentolamine was used to define the nonspecific binding. Clonidine was used as the ligand to define α-adrenergic binding affinity to simplify the comparison with previous results.

Results and Discussion

We note three general trends from comparing the biochemical data of **9** and **16**–**21** (Table 2) with those of **8** and **10**–**15** (Table 1). First, **9** and **16**–**21** are more potent than their corresponding fluoromethyl analogues (**8**, **10**–**15**). Second, while **9** and **16**–**21** show increased affinity for the α_2-adrenoceptor compared to their corresponding fluoromethyl analogues (**8**, **10**–**15**), they are still up to 6-fold more selective than **8** and **10**–**15**. Third, the PNMT inhibitory potencies of **16**–**19** and **10**–**13** followed a similar trend compared to their parent **9** or **8**, respectively, whereby the *N*-substituted compounds were less potent than their parent. The exceptions are *N*-trifluoroethyl sulfonamide **20** and *N*-trifluoropropyl sulfonamide **21**, which are twice as potent as unsubstituted sulfonamide **9**. This is a different trend than that observed for the addition of a 2,2,2-trifluoroethyl or a 3,3,3-trifluoropropyl substituent to the sulfonamide nitrogen of **8**, whereby the addition of these substituents did not increase PNMT inhibitory potency over the parent compound (**8**).[22]

Molecular modeling studies were performed to investigate the differences in PNMT inhibitory potencies between 3-hydroxymethyl-7-(*N*-substituted aminosulfonyl)-THIQs (**9**, **16**–**21**) and their corresponding fluoromethyl analogues (**8**, **10**–**15**). The X-ray crystal structure of PNMT[27] cocrystallized with AdoHcy (**4**) and SK&F 29661 (**5**) has been reported. This crystal structure was used for the docking (AutoDock 3.0)[28] studies of these inhibitors. The docking of inhibitors into the PNMT active site in this paper was performed on the *R*-enantiomer, since previous studies on 3-fluoromethyl- and 3-hydroxymethyl-7-substituted-THIQs indicated that the *R*-enantiomer of these 3-substituted-THIQs is preferred over the *S*-enantiomer in the hPNMT active site.[29,30] Docking of 3-hydroxymethyl-7-(*N*-substituted aminosulfonyl)-THIQs (Figure 3 shows **20**) into the PNMT active site indicates that the hydroxy moiety appears to form a hydrogen bond with Glu219. In contrast, docking studies on 3-fluoromethyl-7-(*N*-substituted aminosulfonyl)-THIQs (Figure 4 shows **14**) indicated that the 3-fluoromethyl moiety appears to make a hydrophobic contact with Tyr222.[22] The docking studies also show that the substituent on the sulfonamide nitrogen of the 3-hydroxymethyl compounds appears to bind in the auxiliary binding pocket in a manner similar to that of their fluoromethyl analogue[22] (Figures 3 and 4). Hydrophobic contacts appear to be made between the sulfonamide substituent and Val53 and Arg44. These docking studies are consistent with the biochemical data that suggest that 3-hydroxymethyl-7-(*N*-substituted aminosulfonyl)-THIQs (**9**, **16**–**21**; Table 2) are more potent than their corresponding 3-fluoromethyl analogues (**8**, **10**–**15**; Table 1), since a

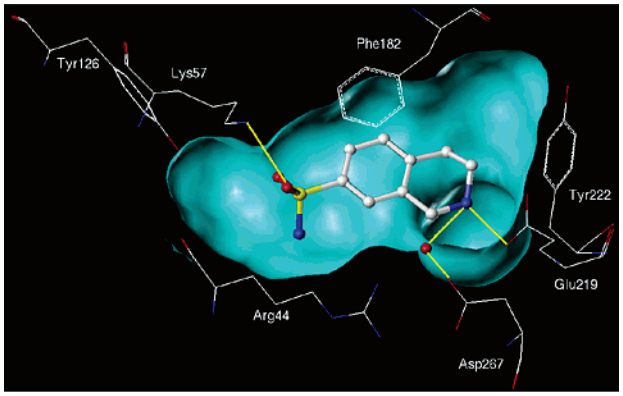


Figure 2. This figure shows the amino acid residues that could interact with SK&F 29661 (**5**) within the hPNMT[27] active site. A Connolly (solvent accessible) surface of the active site revealing SK&F 29661 (**5**) is also shown. Yellow lines indicate possible hydrogen bonds. Carbon is white, nitrogen is blue, oxygen is red, and sulfur is yellow. Hydrogens are not shown for clarity.

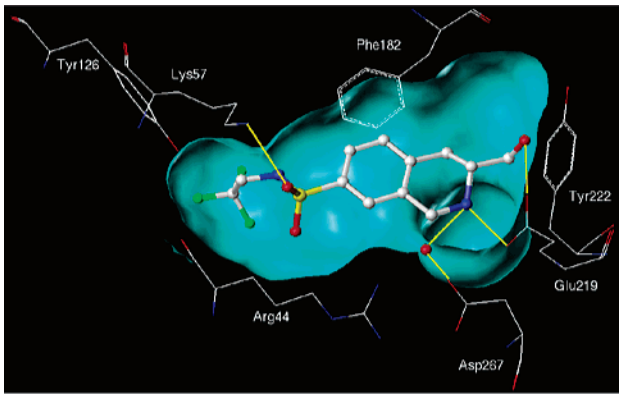


Figure 3. This figure shows **20** docked into the active site of hPNMT[27] and the amino acid residues that could interact with **20**. A Connolly (solvent accessible) surface of the active site revealing **20** is also shown. The docking indicates how the 3-hydroxymethyl group may interact with Glu219. Yellow lines indicate possible hydrogen bonds. Carbon is white, nitrogen is blue, fluorine is green, oxygen is red, and sulfur is yellow. Hydrogens are not shown for clarity.

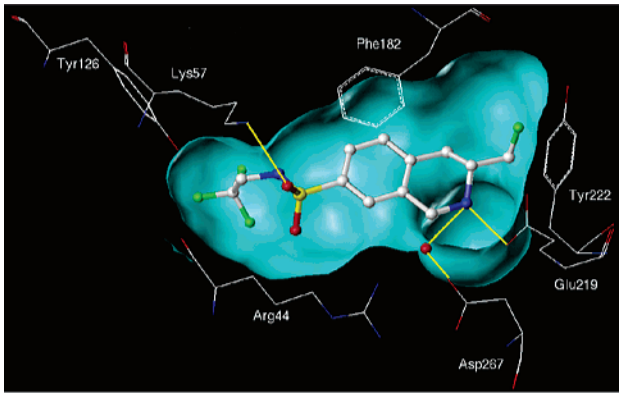


Figure 4. This figure shows **14** docked into the active site of hPNMT and the amino acid residues that could interact with **14**. A Connolly (solvent accessible) surface of the active site revealing **14** is also shown. The docking indicates how the 3-fluoromethyl group may interact with Tyr222. Yellow lines indicate possible hydrogen bonds. Carbon is white, nitrogen is blue, fluorine is green, oxygen is red, and sulfur is yellow. Hydrogens are not shown for clarity.

hydrogen bond results in a significant gain in energy (ca. 3−5 kcal/mol) compared to a hydrophobic contact. For example, *N*-trifluoroethyl sulfonamide **20** (3-CH_2-OH) is approximately six times more potent at inhibiting PNMT than its 3-fluoromethyl analogue **14**. Verification of these interactions will require crystal structures of hPNMT cocrystallized with a 3-fluoromethyl-7-substituted-THIQ and a similarly 7-substituted 3-hydroxymethyl-THIQ. It is not readily apparent from the modeling why *N*-trifluoroethyl sulfonamide **20** (Figure 3) and *N*-trifluoropropyl sulfonamide **21** are more potent than parent **9** (3-CH_2OH-7-SO_2NH_2) nor why *N*-trifluoroethyl sulfonamide **14** (Figure 4) and *N*-trifluoropropyl sulfonamide **15** are equipotent to parent **8** (3-CH_2F-7-SO_2NH_2). However, the biochemical data suggest that the 3-hydroxymethyl group and 7-trifluoroalkyl aminosulfonyl substituent of *N*-trifluoroethyl sulfonamide **20** and *N*-trifluoropropyl sulfonamide **21** are highly favored in the PNMT active site.

In conclusion, the addition of nonpolar substituents to the sulfonamide nitrogen of 7-aminosulfonyl-3-hydroxymethyl-THIQ (**9**) has produced PNMT inhibitors that are more lipophilic than parent **9** and that retained excellent PNMT inhibitory potency and selectivity. Compounds **20** (PNMT K_i = 23 nM) and **21** (PNMT K_i = 28 nM) are the most potent and selective (α_2 K_i/PNMT $K_i \geq$ 15 000) PNMT inhibitors reported to date that, on the basis of their calculated log *P* values, should have significant penetration into the CNS. These compounds may prove useful as pharmacological tools to elucidate the role of CNS Epi.

Experimental Section

All of the reagents and solvents used were reagent grade or were purified by standard methods before use. Melting points were determined in open capillary tubes on a Thomas-Hoover melting point apparatus calibrated with known compounds but are otherwise uncorrected. All proton (^{1}H NMR) and carbon (^{13}C NMR) nuclear magnetic resonance spectra were taken on a Bruker DRX-400 or a Bruker AM-500 spectrometer. Unless otherwise noted, all spectra were collected on the Bruker DRX-400. High-resolution mass spectra (HRMS) were obtained on a VG Analytical ZAB. Elemental analyses were performed by Quantitative Technologies, Inc. (Whitehouse, NJ). Flash chromatography was performed using silica gel 60 (230−400 mesh) supplied by Universal Adsorbents, Atlanta, GA. Anhydrous tetrahydrofuran (THF) and diethyl ether (Et_2O) were distilled from sodium−benzophenone ketyl. Hexanes refers to the mixture of hexane isomers (bp 40−70 °C). All reactions that required anhydrous conditions were performed under argon, and all glassware was either oven-dried or flame-dried before use. (±)-1,2,3,4-Tetrahydro-3-isoquinoline carboxylic acid hydrochloride (**22**) was obtained from Sigma-Aldrich (Milwaukee, WI). AdoMet was obtained from Sigma-Aldrich (St. Louis, MO). [*methyl*-^{3}H]AdoMet and [^{3}H]clonidine were obtained from Perkin-Elmer (Boston, MA).

Radiochemical Assay of PNMT Inhibitors. The assay used in this study has been modified from that described previously.[25] A typical assay mixture consisted of 25 μL of 0.5 M phosphate buffer (pH 8.0), 25 μL of 50 μM unlabeled AdoMet, 5 μL of [*methyl*-^{3}H]AdoMet, containing approximately 3×10^5 dpm (specific activity approximately 15 Ci/mmol), 25 μL of substrate solution (phenylethanolamine), 25 μL of inhibitor solution, 25 μL of enzyme preparation (containing 30 ng hPNMT and 25 μg of bovine serum albumin), and sufficient water to achieve a final volume of 250 μL. After incubation for 30 min at 37 °C, the reaction mixture was quenched by addition of 250 μL of 0.5 M borate buffer (pH 10.0) and was extracted with 2 mL of toluene/isoamyl alcohol (7:3). A 1 mL portion of the organic layer was removed, transferred to a scintillation vial, and diluted with cocktail for counting. The mode of inhibition was ascertained to be competitive in all cases reported in Tables 1 and 2 by examination of the correlation coefficients (r^2) for the fit routines as calculated in the Enzyme Kinetics module (version 1.1) in SigmaPlot (version 7.0).[31] While all K_i values reported were calculated using competitive kinetics, it should be noted that there was not always a great difference between the r^2 values for the competitive model versus the noncompetitive model. All assays were run in duplicate with three inhibitor concentrations over a 5-fold range. K_i values were determined by a hyperbolic fit of the data using the Single Substrate−Single Inhibitor routine in the Enzyme Kinetics module (version 1.1) in SigmaPlot (version 7.0). For inhibitors with apparent IC_{50} values less than 0.1 μM (as determined by a preliminary screen of the compounds to be assayed), the Enzyme Kinetics Tight Binding Inhibition routine was used to calculate the K_i values.

α_2-Adrenoceptor Radioligand Binding Assay. The radioligand receptor binding assay was performed according to the method of U'Prichard et al.[26] Male Sprague−Dawley rats were decapitated, and the cortexes were dissected out and homogenized in 20 volumes (w/v) of ice-cold 50 mM Tris/HCl buffer (pH 7.7 at 25 °C). Homogenates were centrifuged three times for 10 min at 50 000*g* with resuspension of the pellet in fresh buffer between spins. The final pellet was homogenized in 200 volumes (w/v) of ice-cold 50 mM Tris/HCl buffer (pH 7.7 at 25 °C). Incubation tubes containing [^{3}H]clonidine (specific activity approximately 55 Ci/mmol, final concentration 2.0 nM), various concentrations of drugs, and an aliquot of freshly resuspended tissue (800 μL) in a final volume of 1 mL were used. Tubes were incubated at 25 °C for 30 min, and the incubation was terminated by rapid filtration under vacuum through GF/B glass fiber filters. The filters were rinsed with three 5-mL washes of ice-cold 50 mM Tris buffer (pH 7.7 at 25 °C). The filters were counted in vials containing premixed scintillation cocktail. Nonspecific binding was defined as the concentration of bound ligand in the presence of 2 μM of phentolamine. All assays were run in quadruplicate with five inhibitor concentrations over a 16-fold range. IC_{50} values were determined by a log-probit analysis of the data and K_i values were determined by the equation $K_i = IC_{50}/(1 + [\text{clonidine}]/K_D)$, as all Hill coefficients were approximately equal to 1.

Molecular Modeling. Calculated log *P* values and Connolly surfaces were generated in SYBYL on a Silicon Graphics Octane workstation.[20] Docking of the various inhibitors into the PNMT active site was performed using AutoDock 3.0.[28] The default settings for AutoDock were used. The compound

to be docked was initially overlayed with cocrystallized ligand SK&F 29661 (**5**) and minimized with the Tripos force field.

(±)-2-Acetyl-1,2,3,4-tetrahydro-3-isoquinolinecarboxylic Acid (23). A similar procedure was followed as that described by Shinkai et al.[23] Amino acid **22** (1.00 g, 4.67 mmol) was added to a solution of acetone (20 mL) and 2 N NaOH (15 mL). Acetyl chloride (0.801 mL, 9.35 mmol) and 2 N NaOH were added simultaneously slowly at room temperature. The solution was maintained at a pH greater than 10 and stirred for 1 h. The acetone was evaporated in vacuo and the remaining solution was made acidic with 3 N HCl. The aqueous layer was extracted with EtOAc (3 × 50 mL). The combined organic layers were washed with brine and dried over $NaSO_4$. The solution was concentrated in vacuo to yield a light yellow solid (**23**), which needed no further purification (923 mg, 90%): mp 170−171 °C (lit.[32] mp 171−173); ^{1}H NMR (400 MHz, DMSO-d_6) δ 7.28−7.13 (m, 4H), 5.39−5.37 (m, 1H), 4.66 (s, 2H), 3.34−3.29 (m, 1H), 3.15−3.10 (m, 1H), 2.28 (s, 3H); HRMS (FAB+) *m/z* calcd for $C_{12}H_{14}NO_3$ (MH^+) 220.0974 obsd 220.1231.

(±)-2-Acetyl-3-(1-piperidinocarbonyl)-1,2,3,4-tetrahydroisoquinoline (24). Compound **23** (228 mg, 1.04 mmol), 1-hydroxybenzotriazole (155 mg, 1.14 mmol), and *N*-methylmorpholine (0.23 mL, 2.08 mmol) were dissolved in dichloromethane (5 mL) and cooled to 0 °C. Benzotriazol-1-yloxytris-(dimethylamino)phosphonium hexafluorophosphate (506 mg, 1.14 mmol) was added to the solution and the mixture stirred for 15 min. Piperidine (0.134 mL, 1.35 mmol) was added and the solution was stirred at room temperature overnight. The solution was washed with 3 N HCl and brine and dried over Na_2SO_4. The solution was concentrated in vacuo to give a light brown oil. The oil was subsequently purified by flash chromatography eluting with EtOAC → 95:5 EtOAc:MeOH to yield **24** as a clear foamy oil (308 mg, 98%): ^{1}H NMR (500 MHz, $CDCl_3$) δ 7.22−7.13 (m, 4H), 5.55 (m, 1H), 4.69 (m, 2H), 3.60−3.37 (m, 4H), 3.06 (m, 2H), 2.24 (s, 3H), 1.67−1.27 (m, 6H); HRMS (FAB+) *m/z* calcd for $C_{17}H_{23}N_2O_2$ (MH^+) 287.1760 obsd 287.1764.

(±)-2-Acetyl-3-(1-piperidinocarbonyl)-7-sulfonyl chloride-1,2,3,4-tetrahydroisoquinoline (25). Compound **24** (8.24 g, 28.8 mmol) was dissolved in chlorosulfonic acid (100 mL) and stirred at room temperature overnight. The reaction mixture was carefully pipetted onto ice. The mixture was extracted with ethyl acetate (3 × 75 mL), and the combined organic layers were washed with brine and dried over sodium sulfate. The solution was concentrated in vacuo to give a yellow foamy solid. The two regioisomers **25** and **26** were separated by flash chromatography using a gradient of 40:60 hexanes:EtOAc → 30:70 hexanes:EtOAc → 20:80 hexanes:EtOAc → 10:90 hexanes:EtOAc → 5:95 hexanes:EtOAc → EtOAc. The desired 7-sulfonyl chloride (**25**) eluted off the column first as a light yellow oily foam (4.87 g, 44%), followed by the 6-sulfonyl regioisomer (**26**) also as a light oily foam (750 mg, 15%). Compound **25** data: ^{1}H NMR (500 MHz, $CDCl_3$) δ 7.88 (m, 1H), 7.77 (s, 1H), 7.44 (d, J = 8.2 Hz, 1H), 5.83 (m, 1H), 4.83−4.71 (m, 2H), 3.68−3.59 (m, 2H), 3.45−3.33 (m, 2H), 3.22−3.11 (m, 2H), 2.26 (s, 3H), 1.71−1.53 (m, 6H); HRMS (FAB+) *m/z* calcd for $C_{17}H_{22}N_2O_4SCl$ (MH^+) 385.0989 obsd 385.0971.

General Procedure for 27−32. Sulfonyl chloride **25** (500 mg, 1.30 mmol) was dissolved in EtOAc (25 mL) and sat. $NaHCO_3$ (10 mL). The requisite amine was added and the biphasic solution was stirred for 12 h. The organic phase was separated and the aqueous phase was saturated with sodium chloride. The aqueous phase was extracted with EtOAc. The combined organic layers were washed with 3 N HCl that was saturated with sodium chloride, washed with brine, and dried over sodium sulfate. After concentration in vacuo, the crude oily foam was purified by flash chromatography eluting with a gradient of 10:1 EtOAc:hexanes → EtOAc.

(±)-2-Acetyl-7-(*N*-ethylaminosulfonyl)-3-(1-piperidinocarbonyl)-1,2,3,4-tetrahydroisoquinoline (27). Ethylamine hydrochloride (213 mg, 3.90 mmol) was used to give **27** as a oily foamy solid (335 mg, 65%): ^{1}H NMR (MeOH-d_4) δ 7.74−7.67 (m, 2H), 7.43 (d, J = 7.7 Hz, 1H), 5.49 (m, 1H), 4.94−4.72 (m, 2H), 3.70−3.58 (m, 4H), 3.27−3.22 (m, 2H), 3.12−3.08 (m, 2H), 3.01−2.98 (m, 1H), 2.93−2.88 (m, 1H), 2.27 (s, 3H), 1.73−1.46 (m, 4H), 1.08 (t, J = 7.2 Hz, 3H); HRMS (FAB+) *m/z* calcd for $C_{19}H_{28}N_3O_4S$ (MH^+) 394.1801 obsd 394.1802.

(±)-2-Acetyl-3-(1-piperidinocarbonyl)-7-(*N*-propylaminosulfonyl)-1,2,3,4-tetrahydroisoquinoline (28). *n*-Propylamine (0.32 mL, 3.90 mmol) was used to yield **28** as an oily foamy solid (300 mg, 57%): ^{1}H NMR (500 MHz, DMSO-d_6) δ 7.63 (ex s, 1H), 7.58 (d, J = 7.4 Hz, 1H), 7.53 (m, 1H), 7.40 (d, J = 8.0 Hz, 1H), 5.53 (m, 1H), 4.91−4.88 (m, 1H), 4.59−4.55 (m, 1H), 3.52−3.44 (m, 4H), 3.05−2.96 (m, 2H), 2.73−2.51 (m, 2H), 2.15 (s, 3H), 1.61−1.54 (m, 4H), 1.42−1.37 (m, 4H), 0.81 (t, J = 7.3 Hz, 3H); HRMS (FAB+) *m/z* calcd for $C_{20}H_{30}N_3O_4S$ (MH^+) 408.1957 obsd 408.1957.

(±)-2-Acetyl-3-(1-piperidinocarbonyl)-7-(*N*-thiomorpholinosulfonyl)-1,2,3,4-tetrahydroisoquinoline (29). Thiomorpholine (0.39 mL, 3.88 mmol) was used to yield **29** as an oily foamy solid (322 mg, 55%): ^{1}H NMR (500 MHz, DMSO-d_6) δ 7.70 (s, 1H), 7.62 (m, 1H), 7.46 (m, 1H), 5.53 (m, 1H), 4.95−4.92 (m, 1H), 4.60−4.57 (m, 1H), 3.52−3.44 (m, 4H), 3.17 (m, 4H), 3.08−3.03 (m, 2H), 2.69−2.66 (m, 4H), 2.15 (s, 3H), 1.61−1.39 (m, 6H); HRMS (FAB+) *m/z* calcd for $C_{21}H_{30}N_3O_4S$ (MH^+) 452.1678 obsd 452.1665.

(±)-2-Acetyl-7-(*N*-4-chlorophenylaminosulfonyl)-3-(1-piperidinocarbonyl)-1,2,3,4-tetrahydroisoquinoline (30). 4-Chloroaniline (500 mg, 3.90 mmol) and pyridine (5 mL) were used to yield **30** as an oily foamy solid (360 mg, 58%): ^{1}H NMR ($CDCl_3$) δ 7.66 (ex s, 1H), 7.62 (d, J = 7.4 Hz, 1H), 7.35 (m, 2H), 7.23−7.20 (m, 2H), 7.09−7.05 (m, 2H), 5.46 (m, 1H), 4.91−4.65 (m, 2H), 3.65−3.52 (m, 4H), 3.22−3.03 (m, 2H), 2.23 (s, 3H), 1.72−1.52 (m, 6H); HRMS (FAB+) *m/z* calcd for $C_{23}H_{27}N_3O_4SCl$ (MH^+) 476.1411 obsd 476.1384.

(±)-2-Acetyl-3-(1-piperidinocarbonyl)-7-(*N*-2,2,2-trifluoroethylaminosulfonyl)-1,2,3,4-tetrahydroisoquinoline (31). 2,2,2-Trifluoroethylamine (0.310 mL, 3.90 mmol) and pyridine (2 mL) were used to yield **31** as an oily foamy solid (357 mg, 61%): ^{1}H NMR (500 MHz, $CDCl_3$) δ 7.64−7.61 (m, 2H), 7.30 (m, 1H), 6.50 (ex s, 1H), 5.70 (m, 1H), 4.72 (m, 2H), 3.54−3.39 (m, 5H), 3.34 (m, 1H), 3.12 (m, 2H), 2.22 (s, 3H), 1.65−1.62 (m, 6H); HRMS (FAB+) *m/z* calcd for $C_{19}H_{25}N_3O_4F_3S$ (MH^+) 448.1518 obsd 448.1526.

(±)-2-Acetyl-3-(1-piperidinocarbonyl)-7-(*N*-3,3,3-trifluoropropylaminosulfonyl)-1,2,3,4-tetrahydroisoquinoline (32). 3,3,3-Trifluoropropylamine hydrochloride (291 mg, 1.95 mmol) and pyridine (2 mL) were used to yield **32** as an oily foamy solid (386 mg, 64%): ^{1}H NMR (400 MHz, MeOH-d_4) δ 7.76−7.71 (m, 2H), 7.44 (m, 1H), 5.52 (m, 1H), 4.90−4.77 (m, 2H), 3.69−3.22 (m, 6H), 3.11 (m, 2H), 2.42−2.36 (m, 2H), 2.31 (s, 3H), 1.84−1.53 (m, 6H); HRMS (FAB+) *m/z* calcd for $C_{20}H_{27}N_3O_4F_3S$ (MH^+) 462.1674 obsd 462.1659.

General Procedure for 16−19 (selected procedure for **16**). Compound **27** (335 mg, 0.852 mmol) was dissolved in 6 N HCl (3 mL) and heated to reflux for 12 h. The solution was concentrated and evaporated to complete dryness to yield **33**·HCl. Dry THF (15 mL) and 1 M BH_3·THF (3.40 mL, 2.66 mmol) were added, and the mixture was heated to reflux for 12 h. The solution was cooled with an ice bath and quenched by the slow addition of MeOH until bubbling ceased. The solution was concentrated in vacuo, and MeOH (20 mL) and 6 N HCl (5 mL) were added. The reaction mixture was heated to reflux for 3 h. The MeOH was removed and the solution was made basic with 10 N NaOH. The solution was saturated with NaCl and extracted with EtOAc (3 × 50 mL). The organic layers were combined, washed with brine, and dried over sodium sulfate. The solution was concentrated in vacuo and the crude oil was purified by flash chromatography. The purified free amine was converted to its HCl salt by dissolving it in MeOH and bubbling HCl(g) through the solution.

(±)-3-Hydroxymethyl-7-(*N*-ethylaminosulfonyl)-1,2,3,4-tetrahydroisoquinoline Hydrochloride (16·HCl). The crude free amine was purified by flash chromatography eluting with

10:1 EtOAc:MeOH. The hydrochloride salt was recrystallized from EtOH/hexanes to yield **16**·HCl as white crystals (49 mg, 19%): mp 249−250 °C; ^{1}H NMR (500 MHz, DMSO-d_6) δ 9.55 (br ex s, 2H), 7.71−7.47 (m, 3H), 7.45 (d, J = 8.3 Hz, 1H), 5.57 (ex s, 1H), 4.38 (s, 2H), 3.81−3.44 (m, 3H), 3.08−2.98 (m, 2H), 2.79−2.75 (m, 2H), 0.99 (t, J = 7.2 MHz, 3H); ^{13}C NMR (400 MHz, DMSO-d_6) δ 139.7, 137.4, 130.8, 130.7, 126.2, 125.8, 61.3, 54.7, 44.7, 38.4, 28.2, 15.6; HRMS (FAB+) *m/z* calcd for $C_{12}H_{19}N_2O_3S$ (MH^+) 271.1116 obsd 271.1102. Anal. ($C_{12}H_{18}N_2O_3S$·HCl) C, H, N.

(±)-3-Hydroxymethyl-7-(*N*-propylaminosulfonyl)-1,2,3,4-tetrahydroisoquinoline Hydrochloride (17·HCl). Compound **28** (280 mg, 0.912 mmol) and 1 M BH_3·THF (2.74 mL, 2.74 mmol) were used. The crude free amine was purified by flash chromatography eluting with 10:1 EtOAc:MeOH. The HCl salt was recrystallized from EtOH/hexanes to yield **17**·HCl as white crystals (73 mg, 33%): mp 209−210 °C; ^{1}H NMR (500 MHz, DMSO-d_6) δ 9.48 (br ex m, 2H), 7.71−7.64 (m, 3H), 7.46 (d, J = 8.0 Hz, 1H), 5.58 (ex s, 1H), 4.40 (s, 2H), 3.78−3.55 (m, 3H), 3.03 (m, 2H), 2.69 (m, 2H), 1.39 (m, 2H), 0.80 (t, J = 7.2 MHz, 3H); ^{13}C NMR (500 MHz, DMSO-d_6) δ 139.3, 136.9, 130.4, 130.2, 125.6, 125.3, 60.8, 54.2, 44.7, 44.2, 27.7, 22.8, 11.5; HRMS (FAB+) *m/z* calcd for $C_{13}H_{21}N_2O_3S$ (MH^+) 285.1273 obsd 285.1271. Anal. ($C_{13}H_{20}N_2O_3S$·HCl) C, H, N.

(±)-3-Hydroxymethyl-7-(*N*-thiomorpholinosulfonyl)-1,2,3,4-tetrahydroisoquinoline Hydrochloride (18·HCl). Compound **29** (264 mg, 0.585 mmol) and 1 M BH_3·THF (1.76 mL, 1.76 mmol) were used. The crude free amine was purified by flash chromatography eluting with a gradient of 10:1 EtOAc:MeOH → 10:2 EtOAc:MeOH. The HCl salt was recrystallized from EtOH/hexanes to yield **18**·HCl as white crystals (104 mg, 49%): mp 242−243 °C; ^{1}H NMR (500 MHz, DMSO-d_6) δ 9.48 (br ex s, 2H), 7.70 (s, 1H), 7.61 (m, 1H), 7.50 (m, 1H), 5.55 (ex s, 1H), 4.40 (m, 2H), 3.78 (m, 1H), 3.65 (m, 1H), 3.53 (m, 1H), 3.21 (m, 4H), 3.10−3.03 (m, 2H), 2.68 (m, 4H); ^{13}C NMR (400 MHz, DMSO-d_6) δ 138.6, 135.0, 131.7, 130.9, 126.9, 126.5, 61.4, 54.7, 48.7, 44.9, 28.4, 27.3; HRMS (FAB+) *m/z* calcd for $C_{14}H_{21}N_2O_3S_2$ (MH^+) 329.0994 obsd 329.0975. Anal. ($C_{14}H_{20}N_2O_3S_2$·HCl) C, H, N.

(±)-3-Hydroxymethyl-7-(*N*-4-chlorophenylaminosulfonyl)-1,2,3,4-tetrahydroisoquinoline Hydrochloride (19·HCl). Compound **30** (360 mg, 0.758 mmol) and 1 M BH_3·THF (2.27 mL, 2.27 mmol) were used. The crude free amine was purified by flash chromatography eluting with a gradient of 10:1 EtOAc:MeOH → 10:2 EtOAc:MeOH. The HCl salt was recrystallized from EtOH/hexanes to yield **19**·HCl as white crystals (36 mg, 12%): mp 219−220 °C; ^{1}H NMR (500 MHz, DMSO-d_6) δ 9.44 (br ex s, 2H), 7.72 (s, 1H), 7.63 (d, J = 7.7 Hz, 1H), 7.42 (d, J = 8.0 Hz, 1H), 7.31 (d, J = 8.6 Hz, 2H), 7.13 (d, J = 8.6 Hz, 2H), 5.51 (ex s, 1H), 4.33 (m, 2H), 3.75 (m, 1H), 3.59−3.50 (m, 2H), 3.03−2.90 (m, 2H); ^{13}C NMR (400 MHz, DMSO-d_6) δ 138.5, 138.3, 137.5, 131.4, 130.9, 130.1, 128.9, 126.2, 126.0, 122.2, 61.4, 54.6, 44.9, 28.4; HRMS (FAB+) *m/z* calcd for $C_{16}H_{18}N_2O_3SCl$ (MH^+) 353.0727 obsd 353.0712. Anal. ($C_{16}H_{17}N_2O_3SCl$·HCl) C, H, N.

(±)-7-(*N*-2,2,2-Trifluoroethylaminosulfonyl)-1,2,3,4-tetrahydro-3-isoquinolinecarboxylic Acid Methyl Ester (39). Compound **37** (286 mg, 0.640 mmol) was dissolved in 6 N HCl (3 mL) and heated to reflux for 12 h. The solution was concentrated and evaporated to dryness. The crude amino acid (**43**·HCl) was dissolved in MeOH (10 mL), and thionyl chloride (0.122 mL, 0.960 mmol) was carefully added. The solution was heated to reflux for 12 h and the MeOH was subsequently removed in vacuo. Saturated sodium bicarbonate (10 mL) was added and the mixture was extracted with EtOAc (3 × 30 mL). The combined organic layers were washed with brine and dried over sodium sulfate. The solution was concentrated in vacuo to produce a crude solid that was recrystallized from EtOAC/hexanes to yield **39** as light yellow crystals (136 mg, 60%): mp 155−156 °C; ^{1}H NMR (400 MHz, MeOH-d_4) δ 7.66−7.61 (m, 2H), 7.36 (d, J = 8.1 Hz, 1H), 4.18−4.05 (m, 2H), 3.84−3.79 (m, 4H), 3.79−3.57 (m, 2H), 3.23−3.18 (m, 1H) 3.07−3.00 (m, 1H); HRMS (FAB+) *m/z* calcd for $C_{13}H_{16}N_2O_4F_3S$ (MH^+) 353.0783 obsd 353.0755.

(±)-7-(*N*-3,3,3-Trifluoropropylaminosulfonyl)-1,2,3,4-tetrahydro-3-isoquinolinecarboxylic Acid Methyl Ester (40). Compound **40** was prepared in a similar manner as described for **39**, except **38** (330 mg, 0.717 mmol) was used. The crude solid was recrystallized from EtOAc/hexanes to yield **40** as light yellow crystals (128 mg, 49%): mp 124−125 °C; ^{1}H NMR (400 MHz, MeOH-d_4) δ 7.64 (d, J = 7.9 Hz, 1H), 7.61 (s, 2H), 7.38 (d, J = 8.1 Hz, 1H), 4.19−4.06 (m, 2H), 3.84−3.80 (m, 4H), 3.24−3.19 (m, 1H), 3.09−3.01 (m, 3H), 2.44−2.32 (m, 2H); HRMS (FAB+) *m/ z* calcd for $C_{14}H_{18}N_2O_4F_3S$ (MH^+) 367.0939 obsd 367.0949.

(±)-3-Hydroxymethyl-7-(*N*-2,2,2-trifluoroethylaminosulfonyl)-1,2,3,4-tetrahydroisoquinoline (20). Compound **39** (114 mg, 0.324 mmol) was dissolved in THF (15 mL), and 2 M $LiBH_4$ (0.491 mL, 0.972 mmol) was added. The solution was stirred at room temperature for 12 h and quenched with MeOH. The MeOH and THF were removed, and MeOH (20 mL) and 6 N HCl (5 mL) were added. The solution was gently heated to reflux for 3 h. The reaction workup was similar to that described for **16**. The crude free amine was purified by flash chromatography eluting with a gradient of 12:1 DCM:MeOH → 10:1 DCM:MeOH. The free amine was recrystallized from EtOH/hexanes to yield **20** as white crystals (48 mg, 46%): mp 172−173 °C; ^{1}H NMR (500 MHz, DMSO-d_6) δ 8.45 (br ex s, 1H), 7.55−7.52 (m, 2H), 7.15 (d, J = 8.0 Hz, 1H), 4.74 (ex s, 1H), 4.03−3.90 (m, 2H), 3.67−3.62 (m, 2H), 3.46−3.33 (m, 3H), 2.84−2.74 (m, 2H); ^{13}C NMR (500 MHz, DMSO-d_6) δ 140.7, 138.0, 137.8, 130.3, 124.7 (q, J = 277 Hz, CF_3), 124.4, 124.0, 65.2, 54.9, 47.8, 43.8 (q, J = 34 Hz, CF_3CH_2), 31.7; HRMS (FAB+) *m/z* calcd for $C_{12}H_{16}N_2O_3F_3S$ (MH^+) 325.0834 obsd 325.0833. Anal. ($C_{12}H_{15}N_2O_3F_3S$) C, H, N.

(±)-3-Hydroxymethyl-7-(*N*-3,3,3-trifluoropropylaminosulfonyl)-1,2,3,4-tetrahydroisoquinoline (21). Compound **21** was prepared in a similar manner as described for **20**, except that **40** (115 mg, 0.314 mmol) and 2 M $LiBH_4$ (0.471 mL, 0.940 mmol) were used. The crude free amine was purified by flash chromatography eluting with 10:1 DCM:MeOH. The free amine was recrystallized from EtOH/hexanes to yield **21** as white crystals (64 mg, 60%): mp 145−146 °C; ^{1}H NMR (500 MHz, DMSO-d_6) δ 7.77 (br ex s, 1H), 7.53−7.49 (m, 2H), 7.31 (d, J = 8.0 Hz, 1H), 4.79 (ex s, 1H), 4.04−3.91 (m, 2H), 3.46−3.33 (m, 3H), 2.95−2.92 (m, 2H), 2.85−2.74 (m, 2H), 2.47−2.42 (m, 2H); ^{13}C NMR (400 MHz, DMSO-d_6) δ 141.1, 138.3, 137.5, 130.9, 127.3 (q, J = 275 Hz, CF_3), 125.0, 124.6, 65.7, 55.4, 48.3, 36.9, 33.9 (q, J = 26.8 Hz, CF_3CH_2), 32.2; HRMS (FAB+) *m/z* calcd for $C_{13}H_{18}N_2O_3F_3S$ (MH^+) 339.0990 obsd 339.0973. Anal. ($C_{13}H_{17}N_2O_3F_3S$) C, H, N.

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Supporting Information Available: All elemental analyses (C, H, N) for assayed compounds and X-ray crystal structure data for **21**. This material is available free of charge via the Internet at http://pubs.acs.org.

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