Identification of a Potent, Selective, and Orally Active Leukotriene A_4 Hydrolase Inhibitor with Anti-Inflammatory Activity

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LTA_4H is a ubiquitously distributed 69 kDa zinc-containing cytosolic enzyme with both hydrolase and aminopeptidase activity. As a hydrolase, LTA_4H stereospecifically catalyzes the transformation of the unstable epoxide LTA_4 to the diol LTB_4, a potent chemoattractant and activator of neutrophils and a chemoattractant of eosinophils, macrophages, mast cells, and T cells. Inhibiting the formation of LTB_4 is expected to be beneficial in the treatment of inflammatory diseases such as inflammatory bowel disease (IBD), asthma, and atherosclerosis. We developed a pharmacophore model using a known inhibitor manually docked into the active site of LTA_4H to identify a subset of compounds for screening. From this work we identified a series of benzoxazole, benzthiazole, and benzimidazole inhibitors. SAR studies resulted in the identification of several potent inhibitors with an appropriate cross-reactivity profile and excellent PK/PD properties. Our efforts focused on further profiling JNJ 27265732, which showed encouraging efficacy in a disease model relevant to IBD.

Introduction

The proinflammatory mediator leukotriene B_4 (LTB_4[a]) is a potent chemoattractant and activator of neutrophils and a chemoattractant of eosinophils, macrophages, mast cells, and T cells. It is implicated in disorders such as inflammatory bowel disease (IBD),[1,2] rheumatoid arthritis (RA),[3–5] chronic obstructive pulmonary disease (COPD),[6,7] asthma,[8–12] cancer,[13,14] and atherosclerosis.[15–20] The generation of LTB_4 in vivo is regulated by the action of leukotriene A_4 hydrolase (LTA_4H). LTA_4H is a ubiquitously distributed 69 kDa zinc-containing cytosolic enzyme with both hydrolase and aminopeptidase activity, the crystal structure of which was published in 2001.[21] As a hydrolase, it stereospecifically catalyzes the transformation of the unstable epoxide LTA_4 to the diol LTB_4 and is a key enzyme in the arachidonic acid cascade downstream of the initial action of 5-lipoxygenase (5-LO) and FLAP (5-lipoxygenase-activating protein) as shown in Figure 1. LTA_4H also functions as an anion-dependent aminopeptidase, processing arginyl di- and tripeptides with high efficiency. The biological relevance of this activity is unknown.[22,23] It is important to note that LTA_4H-deficient mice have been generated; these mice develop normally and are healthy.[24] Studies of LTA_4H deficient mice in several models of inflammation clearly establish the contribution of LTB_4 in the inflammatory process[24] and strongly support the pursuit of LTA_4H inhibitors as potential treatments for inflammatory diseases.

A 1996 review details work on inhibitors of 5-LO and FLAP, as well as receptor antagonists of LTD_4 and LTB_4.[25] Inhibitors of 5-LO and antagonists of LTD_4 are currently marketed drugs used in the treatment of asthma. A large number of reports suggesting the use of LTB_4 receptor antagonists as potential therapeutics have also appeared, and although several compounds have been evaluated in human clinical trials, no drugs have been marketed to date.

Additionally in 2000, four groups simultaneously identified a second GPCR for LTB_4 that shares a 45.2% homology with the first receptor.[26–29] These two receptors, currently termed BLT1 (high affinity, cloned in 1997, primarily expressed on neutrophils)[30] and BLT2 (low affinity, expressed in the liver, spleen, and peripheral leukocytes),[28] are both anticipated to play a role in the inflammatory process. Recent reports established the presence of BLT1 and BLT2 receptors on human and murine mast cells and demonstrated chemotaxis of mast cells to LTB_4.[31] In order to achieve efficacy in vivo, an antagonist of both receptors may be needed; inhibition of LTA_4H would obviate the need for a dual antagonist.[32]

We also speculate that inhibitors of LTA_4H may have advantages over inhibitors of 5-LO. For example, blockade of 5-LO prevents the formation of LTA_4, which in turn is one source of the lipoxins, LXA_4 and LXB_4 (lipoxygenase interaction products), as shown in Figure 2.[33–36] In contrast, the inhibition of LTA_4H would not hinder the production of the lipoxins because it acts downstream of this branch point in the cascade. Additionally, researchers at Johnson & Johnson PRD in La Jolla have shown that **33r**, vide infra, when tested in murine whole blood and when dosed orally at 30 mpk in a zymosan-induced peritonitis model, selectively inhibited LTB_4 production without affecting cysteinyl leukotriene production and maintained the production of the anti-inflammatory mediator LXA_4.[37] The significance of these findings is related to the role of lipoxins, which are known endogenous anti-inflammatory agents that participate in the resolution process[38] and are postulated to recruit monocytes. In the presence of lipoxins monocytes undergo maturation to macrophages, which clear inflamed areas of necrotic and apoptotic neutrophils. Importantly, the resulting macrophages lack the capacity to cause additional tissue damage, as they cannot generate superoxide ions.[39–41]

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[a] Abbreviations: LTA_4H, leukotriene A_4 hydrolase; LTA_4, leukotriene A_4; LTB_4, leukotriene B_4; IBD, inflammatory bowel disease; PK/PD, pharmacokinetic/pharmacodynamic; RA, rheumatoid arthritis; COPD, chronic obstructive pulmonary disease; 5-LO, 5-lipoxygenase; FLAP, 5-lipoxygenase-activating protein; LTD_4, leukotriene D_4; GPCR, G-protein-coupled receptor; BLT1, B leukotriene 1 receptor; BLT2, B leukotriene 2 receptor; LXA_4, lipoxin A_4; LXB_4, lipoxin B_4; CADD, computer-assisted drug design; TBAF, tetrabutylammonium fluoride; MPO, myeloperoxidase activity; hERG, human ether-a-go-go related gene; SEM-Cl, 2-(trimethylsilyl)ethoxymethyl chloride; CDI, carbonyl diimidazole.

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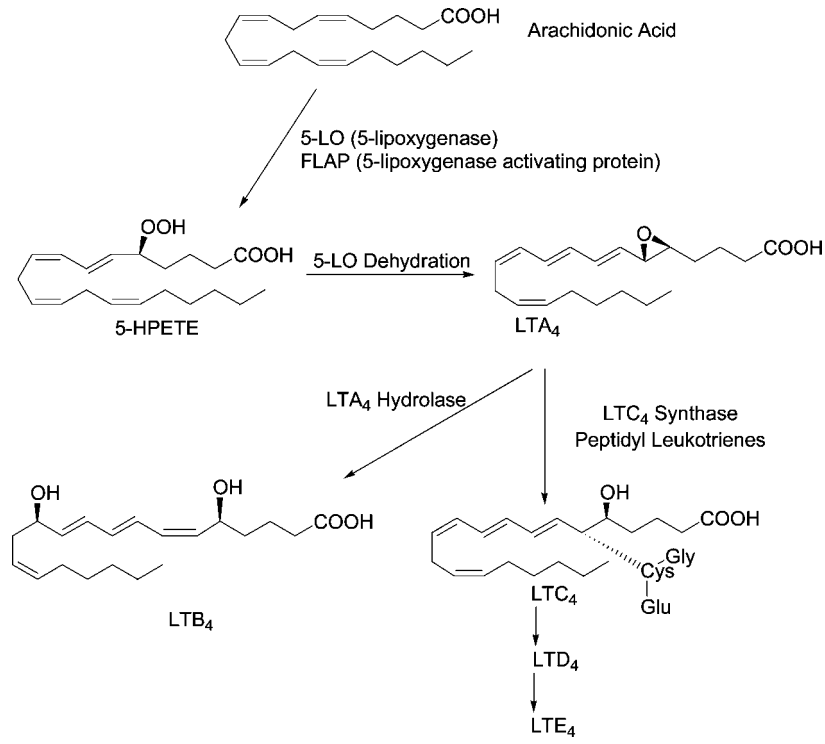


Figure 1. Leukotriene biosynthesis.

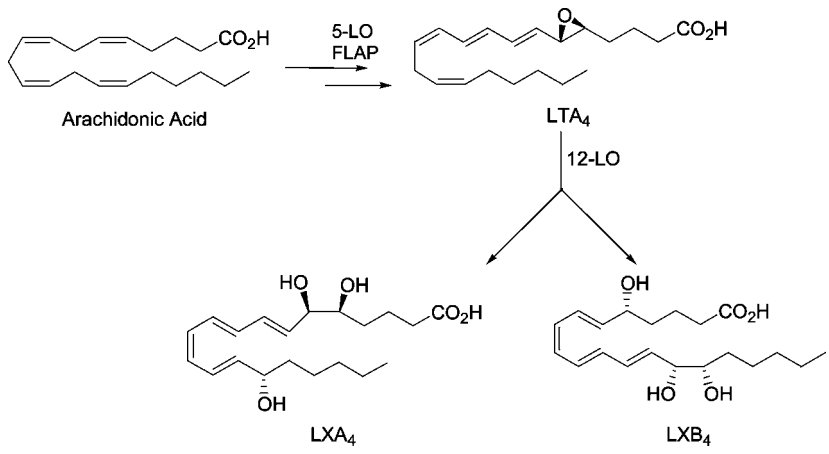


Figure 2. Formation of lipoxins.

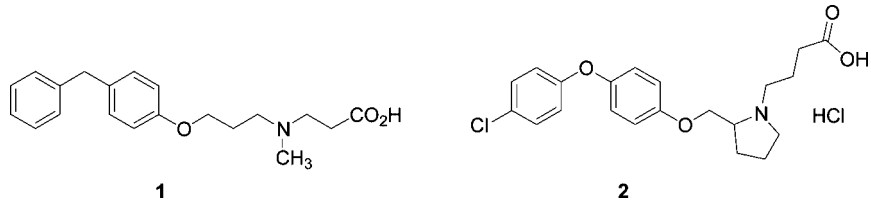


Figure 3. Structures of known LTA_4H inhibitors.

Searle published extensively on their efforts to identify an orally active inhibitor of LTA_4H. The initial work focused on substrate-based and hydroxamic acid dipeptide inhibitors.[42,43] Subsequent work detailed other small molecule inhibitors related to the initial clinical candidate, **1** (SC-57461A) (Figure 3).[44,45] More recent publications disclosed pyrrolidine and piperidine analogues of **1**,[46] as well as imidazopyridines and purines.[47] To date, no reports of an LTA_4H inhibitor advancing to phase I clinical trials from this work have appeared. More recently deCODE Genetics has reported advancing an LTA_4H inhibitor, **2** (DG-051), into phase I and phase II clinical trials for the treatment of cardiovascular disease (Figure 3).[48–50]

Lead Identification and Analogue Design

We disclose our initial efforts to identify orally active inhibitors of LTA_4H. This work focuses on the development of a series of benzoxazoles, benzthiazoles, and benzimidazoles. The La Jolla CADD team developed a pharmacophore model using the Insight II[51] program by which a known inhibitor[44,52] was manually docked into the active site of LTA_4H and

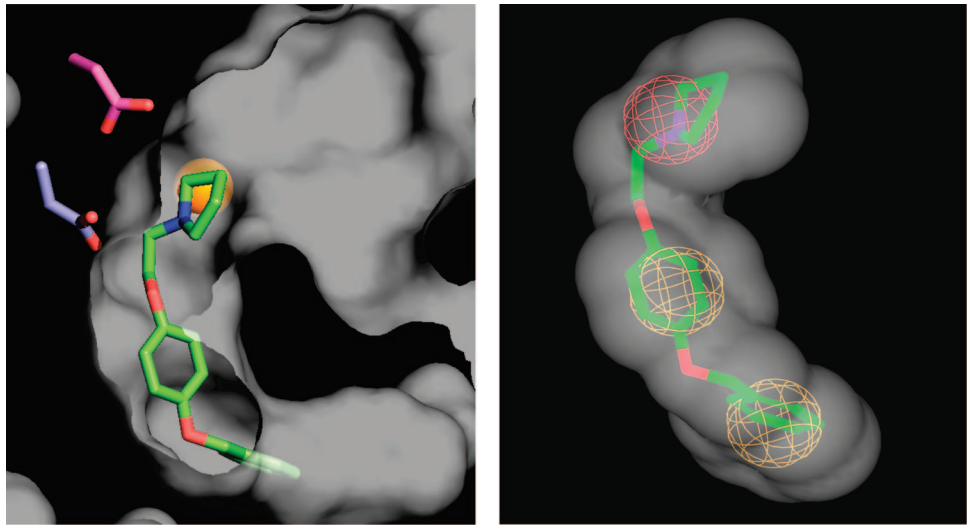
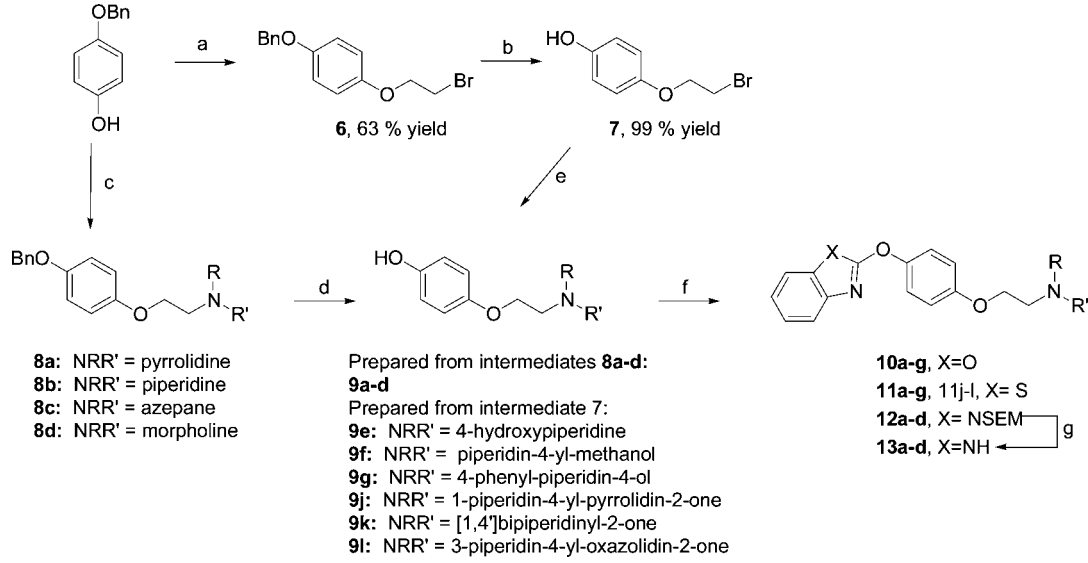


Figure 4. Known inhibitor docked in the active site of LTA_4H (left) and resulting pharmacophore used in virtual screening (right). Graphic was prepared using Pymol v0.99, DeLano Scientific L.L.C.

Scheme 1. Synthesis of Target Compounds Containing the Oxygen Two-Carbon Linker[a]



[a] (a) 1,2-Dibromoethane, K_2CO_3, CH_3CN, Δ; (b) 10% Pd/C, H_2, 40 psi, 10:1 THF/EtOH; (c) 1-(2-chloroethyl)amine, Cs_2CO_3, K_2CO_3, CH_3CN, room temp; (d) 10% Pd/C, H_2, 40 psi, 1:1 EtOH/EtOAc; (e) RR′NH, $^iPr_2EtN$, CH_3CN, room temp. (f) X = O: 2-chlorobenzoxazole, K_2CO_3 or Cs_2CO_3, acetone, 5 °C to room temp. X = S: 2-chlorobenzothiazole, K_2CO_3 or Cs_2CO_3, DMF, Δ. X = NSEM: compound **9**, Cs_2CO_3, DMF, Δ. (g) TBAF, TMEDA, THF, Δ.

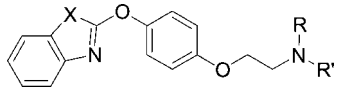
minimized in the presence of the fixed protein. Several conformations of the inhibitor were investigated and minimized as part of the manual docking process. The inhibitor docked in the active site with a Connolly[53] surface representation for the protein and the key residues Glu-271 (in blue) and Glu-296 (in pink) is shown on the left in Figure 3. The potential interaction of the basic amine with these residues suggests a role in inhibitor activity. The resulting pharmacophore, which was constructed using the program Catalyst[54] from one of the lower energy docked conformations, is shown on the right in Figure 3. The key features of the inhibitor emphasized in building the pharmacophore were the basic amine and the two aromatic rings (one for the core and the other for the end fitting into the active site), in addition to molecular shape. As such, the two aromatic features need to be oriented to accommodate the bend in the active site of the enzyme. This model was then used to screen a corporate collection to identify the leads, **3**, **4**, and **5**, shown in Figure 4.

An exploratory chemistry effort focused on the preparation, according to one of the methods illustrated in Scheme 1, of a small set of compounds (Table 1)[55] based on these structures. Investigations to elucidate the SAR further included shortening the linker between the central aromatic ring and the basic nitrogen to address off-target cross-reactivity (not discussed) and potency, as well as changes to the amine component.

Chemistry and Structure−Activity Relationships

Target compounds containing an oxygen two-carbon linker were prepared according to Scheme 1 starting from commercially available 4-benzyloxyphenol. Treatment with dibromoethane at elevated temperature in the presence of K_2CO_3 resulted in a 63% yield of the bromide intermediate **6**. Subsequent removal of the benzyl group of **6** afforded bromophenol **7**. Displacement of the bromide with various amines in the presence of Hünig's base gave intermediates **9e**−**l**. Alternatively, intermediates **9a**−**d** were prepared via reaction

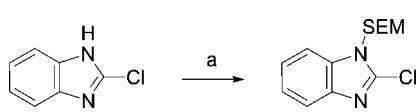
Table 1. In Vitro IC_{50} Enzymatic Data for Initially Prepared Analogues in the Oxygen Two-Carbon Linker Series[54,a]



Compound Number	X =	Amine NRR'	LTA_4H (nM)
10a	O		7*
11a	S		14 ± 9
13a	NH		84 ± 9
10b	O		11 ± 8
11b	S		54 ± 29
13b	NH		110 ± 26
10c	O		4**
11c	S		66 ± 52
13c	NH		140 ± 65
10d	O		58 ± 22
11d	S		350 ± 59
13d	NH		3000 ± 300

[a] Data expressed as IC_{50} ± std dev in nM. (*) Average of 2 determinations. (**) Single determination.

Scheme 2. Preparation of 2-Chloro-1-(2-trimethylsilanylethoxymethyl)-1*H*-benzoimidazole[a]

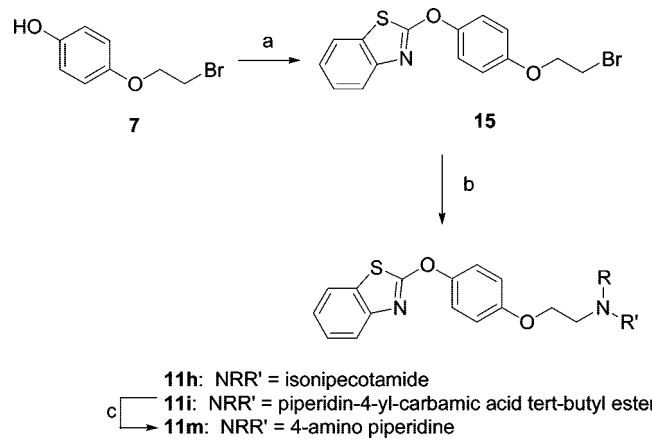


14

[a] (a) (1) NaH, DMF, 5 °C; (2) SEM-Cl, room temp.

of 4-benzyloxyphenol with various 2-chloroethyl amines (**8a**−**d**) in the presence of K_2CO_3 and Cs_2CO_3 followed by removal of the benzyl group.[44] Reaction with 2-chlorobenzoxazole, 2-chlorobenzothiazole, or **14** (prepared according to Scheme 2) gave compounds **10a**−**g**, **11a**−**m**, or **12a**−**d**, respectively. The target benzimidazoles, **13a**−**d**, were obtained after deprotection of intermediates **12a**−**d** using TBAF in the presence of TMEDA at 70 °C.

Scheme 3. Alternative Method for the Synthesis of Compounds Containing the Oxygen Two-Carbon Linker[a]



7 **15**

11h: NRR' = isonipecotamide
11i: NRR' = piperidin-4-yl-carbamic acid tert-butyl ester
11m: NRR' = 4-amino piperidine

[a] (a) 2-Chlorobenzothiazole, Cs_2CO_3, CH_3CN, room temp; (b) RR′NH, silica-bound dimethylamine, CH_3CN, Δ; or RR′NH, K_2CO_3, CH_3CN, Δ; (c) TFA/CH_2Cl_2 (1:1), room temp.

Alternatively, compound **7** when treated with 2-chlorobenzothiazole in the presence of Cs_2CO_3, afforded compound **15** (47% yield), a versatile intermediate for evaluating modifications to the amine functionality as shown in Scheme 3. As an example, treatment of **15** with piperidin-4-yl-carbamic acid *tert*-butyl ester resulted in the formation of **11i**. Conversion to the target compound **11m** was accomplished using a 1:1 mixture of TFA and CH_2Cl_2 at room temperature.

As illustrated in Table 1,[55] these modifications led to compounds with similar potency within both the benzoxazole and benzthiazole series. In general, the benzimidazole analogues were somewhat less active than the corresponding benzoxazoles and benzthiazoles, a trend that is analogous to that observed with the initial hits (Figure 5).

Calculation of the pK_a of the conjugate acid formed by protonation of the amine using Jaguar reveals that the basicity of the morpholine nitrogen of analogues **10d**, **11d**, and **13d** is notably less (7.2) than the corresponding nitrogen of the pyrrolidine/piperidine analogues (8.8) or the azepane (9.6) analogues.[56] The reduced inhibition shown by the morpholine analogues may be correlated to this decrease in basicity resulting in a less optimal binding interaction between the inhibitor and the enzyme, further supporting the docking studies that suggest the protonated amine may be appropriately aligned to make key salt-bridge interactions with the nearby Glu-271 and Glu-296 residues.

Of the molecules listed in Table 1, compound **10b** (JNJ 10392980)[57] provided the desired combination of in vitro enzymatic and mouse whole blood activity ($hLTA_4H$ IC_{50} < 50 nM, MWB LTA_4H IC_{50} < 300 nM) and upon further profiling, it was identified as a potent, orally active inhibitor of LTA_4H. This compound maintained reasonable potency in diluted mouse whole blood with an IC_{50} of 51 and 95 nM using either 1:14 or 1:1 dilution with cell culture media, respectively. Compound **10b** exhibited moderate to high protein binding in human plasma (96%).[58] It showed a clear pharmacokinetic/pharmacodynamic relationship as supported by the efficacy in the murine arachidonic acid induced ear inflammation model when dosed orally at 30 mg/kg. This efficacy was measured by LTB_4 inhibition of ex vivo stimulated mouse whole blood at t = 4 h (postdose) and inhibition of neutrophil influx, as measured by myeloperoxidase activity (MPO). Compound **10b** inhibited both LTB_4 and MPO production by 77% (see Table 6). In vitro

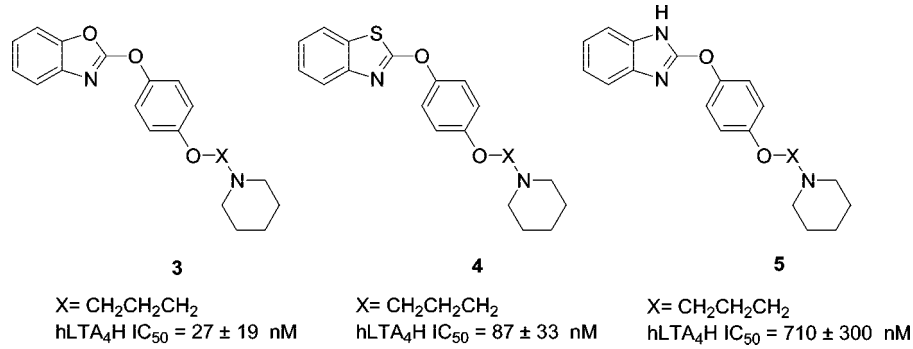
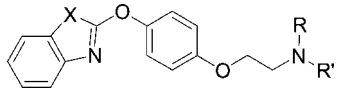


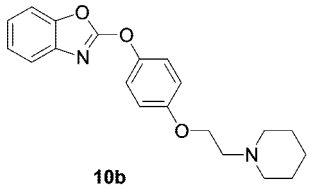
Figure 5. Lead compounds identified through a CADD directed library search.

Table 2. In Vitro IC_{50} Data for Benzoxazole and Benzthiazole Compounds[54,a]



Compound Number	X =	Amine NRR'	LTA_4H (nM)	MWB LTA_4H (nM)
10e	O		9 ± 4	100 ± 68
11e	S		31 ± 8	180 ± 88
10f	O		14 ± 7	46 ±16
11f	S		13 ± 9	100 ± 86
10g	O		6 ± 6	91 ± 30
11g	S		66 ± 33	360 ± 71

[a] Data expressed as IC_{50} ± std dev in nM. Murine whole blood diluted 1:14 with media is stimulated with calcium ionophore, A23187, and then assayed for LTB_4 production.



***In Vitro* Data**
LTA_4H IC_{50} = 11 ± 8 nM
MWB LTA_4H IC_{50} = 51 ± 35 nM
Solubility: pH = 2, NA
pH 7.4 0.16 mg/mL
Stability $t_{1/2}$: pH 2 = 90 min
pH 7.4 >1000 min
Cardiovascular:
hERG AB IC_{50} = 1 μM

Figure 6. Representative data for compound **10b**.

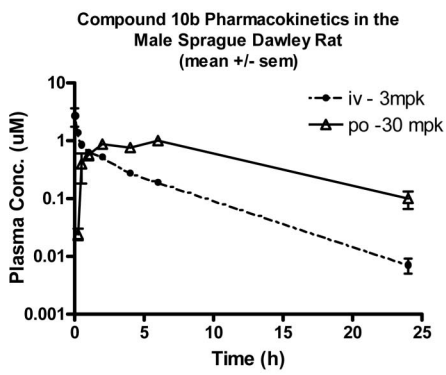
evaluation of metabolic stability indicated that the compound is stable to liver microsomes or S9 fraction across all species evaluated (murine, rat, dog, and human) and does not inhibit any of the major P_{450} isoforms.[59] Evaluation of the compound in a 50-receptor and ion channel panel and a 24-enzyme panel (including enzymes within leukotriene related pathways COX-1/2 and PLA_2) showed minimal cross-reactivity.[37,60,61] However, this compound showed very low stability under acidic conditions, preventing accurate solubility determinations at pH 2.[62] Additionally, the inhibition of the hERG (human ether-a-go-go) channel (Figure 6) as measured by astemizole binding[63] (hERG AB) and the pharmacokinetic profile in dog (Figure 7) of **10b** are less preferred.[64] The hERG binding data for all of the other compounds listed in Table 1 with suitable in vitro activity had IC_{50} values similar to that of **10b** in the hERG AB assay, precluding additional profiling. Replacement of the oxygen connecting the benzoxazole to the central aromatic ring with a methylene resulted in compounds with improved stability at pH 2; however, in general, these analogues were an order of magnitude less active (data not shown). Thus, our efforts to identify pH stable, orally active inhibitors of LTA_4H with minimal cross reactivity continued with a renewed focus on modifications to both the amine portion and the linker length.

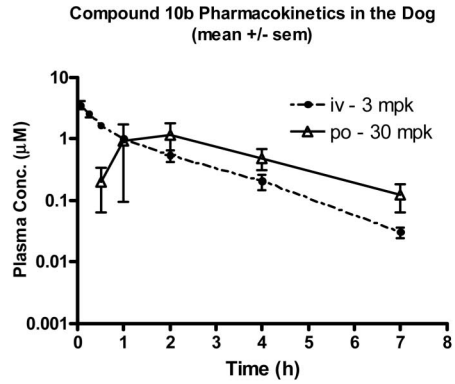
Modification of the amine portion of **10b** was well tolerated in terms of enzyme inhibition of purified enzyme preparations and mouse whole blood as illustrated in Table 2.[55] Similar changes in the benzthiazole series, Tables 2 and 3,[55] were also well tolerated with respect to potency and efficacy (Table 6, entries **11f**, **11h**, and **11j**) and ultimately led to compounds with superior PK properties (data not shown). Additionally, the stability of the benzthiazole compounds at low pH was significantly greater than the benzoxazoles, frequently exceeding 24 h at pH 2, leading us to focus our efforts more fully on this subset of analogues.

The amine, **19**, used in the preparation of target compound **11l** was synthesized according to the procedure shown in Scheme 4. Starting from commercially available 1-benzyl-4-piperidinone, reductive amination with ethanolamine followed by treatment with 1,1′-carbonyldiimidazole (CDI) led to intermediate **18**. Removal of the benzyl protecting group using α-chloroethyl chloroformate led to the desired amine **19** as the HCl salt.

The in vivo efficacy of several of the compounds from Tables 2 and 3 were evaluated at 30 mg/kg, po, in the murine arachidonic acid induced ear inflammation model as shown in Table 6. The efficacy increased significantly for compound **11f** relative to compound **11e** (**11e**, LTB_4 inhibition = 32%, MPO inhibition = 29%; **11f**, LTB_4 inhibition = 78%, MPO inhibition = 83%). In both cases, the addition of the hydroxyl group resulted in a reduction in the astemizole binding relative to the parent piperidine (**10b**, IC_{50} = 1 μM; **11e**, IC_{50} = 12 μM; and **11f**, IC_{50} = 3.6 μM). Entry **11h** was also orally efficacious in the mouse model (LTB_4 inhibition = 79%, MPO inhibition = 79%, at 30 mg/kg, Table 6), but both **11f** and **11h** suffered from poor oral exposure in a rat pharmacokinetic model (data not shown). Compound **11j** was identified as a potent and orally active inhibitor of LTA_4H (LTB_4 inhibition = 67%, MPO inhibition = 84%) with encouraging rat pharmacokinetics (not shown). However, the inhibition of astemizole binding to the hERG channel (IC_{50} = 2.7 μM) exhibited by this compound and the related analogues **11k** and **11l** (IC_{50} = 1.8 and 2.7 μM, respectively) again prompted continued investigation. Additional modifications included shortening of the linker length from oxygen with two-carbon to two-carbon and ultimately to one-carbon as discussed below.

The two-carbon linker analogues were prepared according to the method shown in Scheme 5. Treatment of commercially available 4-hydroxyphenethyl alcohol, **20**, with HBr at 80 °C gave the desired bromide **21** in quantitative yield. Reaction under standard conditions, as detailed above, yielded the amine



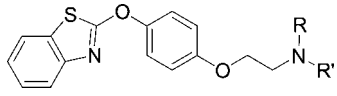


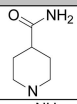
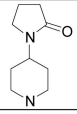
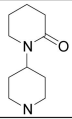
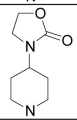
Route	Dose (mg/Kg)	t_{max} (h)	C_{max} (µmol/L)	AUC (h*µmol/L)	CL (L/h)	V_{ss} (L/Kg)	$t_{1/2}$ (h)	%F
Rat iv	3	--	3.8	4.8	0.57	7	2.6	--
Rat po	30	4.6	1.1	15	--	--	5.2[a]	31
Dog iv	3	--	4.5	3.9	0.78	3.2	1	--
Dog po	30	2.3	1.2	4.2	--	--	2.3	11

[a]Half-life is an estimate of the terminal elimination phase from the oral dose.

Figure 7. Pharmacokinetic profile of **10b** in rat and dog.

Table 3. In Vitro IC_{50} Data for Benzthiazole Compounds[54,a]

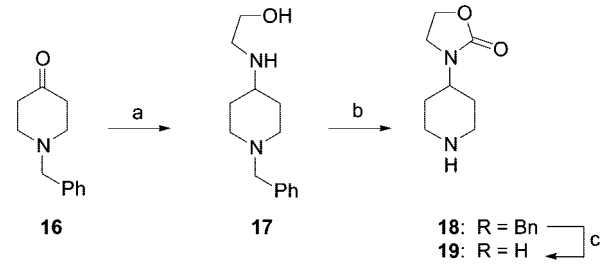


Compound Number	Amine NRR'	LTA_4H (nM)	MWB LTA_4H (nM)
11h		13 ± 5	120 ±11
11m		66 ± 28	430 ±210
11j		16 ± 5	130 ± 77
11k		70 ± 23	170 ± 130
11l		11 ± 2	60 ± 25

[a] Data expressed as IC_{50} ± std dev in nM. Murine whole blood diluted 1:14 with media is stimulated with calcium ionophore, A23187, and then assayed for LTB_4 production.

intermediates **24b** and **24e**. Further reaction with 2-chlorobenzothiazole in the presence of Cs_2CO_3 in DMF at elevated temperature afforded the desired target compounds **27b** and **27e**. Analogously, compound **20** was treated with 2-chlorobenzothiazole directly followed by bromination using PBr_3 to yield **25**.

Scheme 4. Preparation of Amine **15**[a]

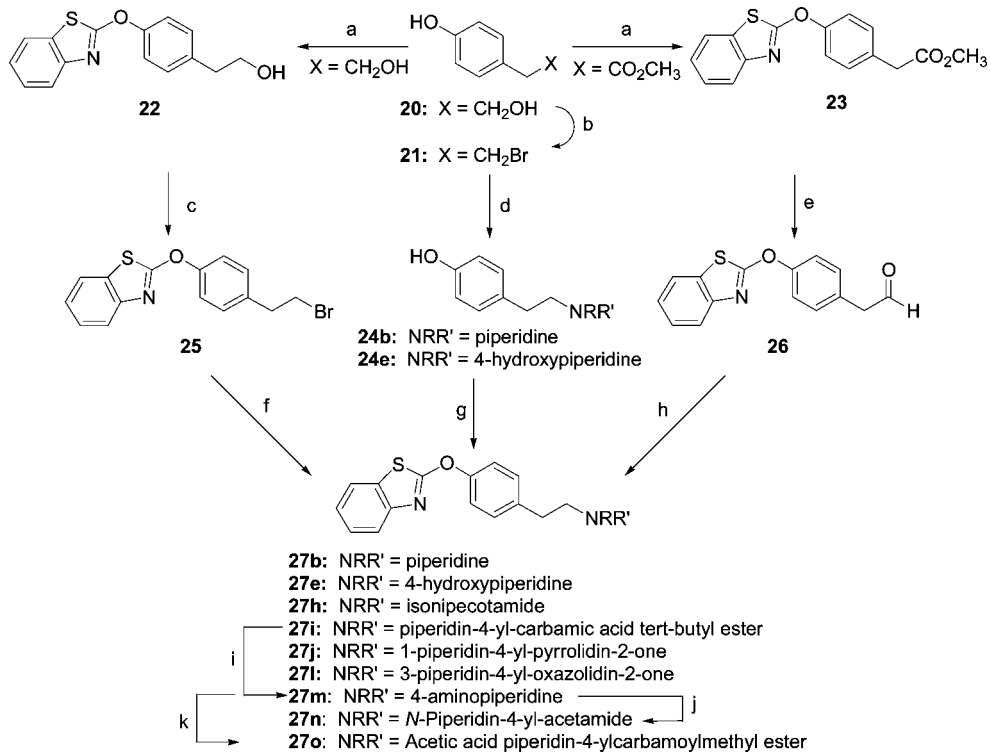


[a] (a) $H_2NCH_2CH_2OH$, $NaCNBH_3$, TfOH, CH_3OH, room temp; (b) CDI, $ClCH_2CH_2Cl$, room temp; (c) α-chloroethyl chloroformate, $ClCH_2CH_2Cl$, 100 °C.

Intermediate **25** was converted to target compounds **27h** and **27j** via alkylation of the appropriate amine. Alternatively, starting with commercially available 4-hydroxyphenylacetic acid methyl ester, intermediate **26** was prepared via alkylation of the hydroxyl group and subsequent reduction of the methyl ester **23** to the aldehyde under carefully controlled reaction conditions. Conversion to target compounds **27i** and **27l** was achieved by reductive amination using the desired amine.[65]

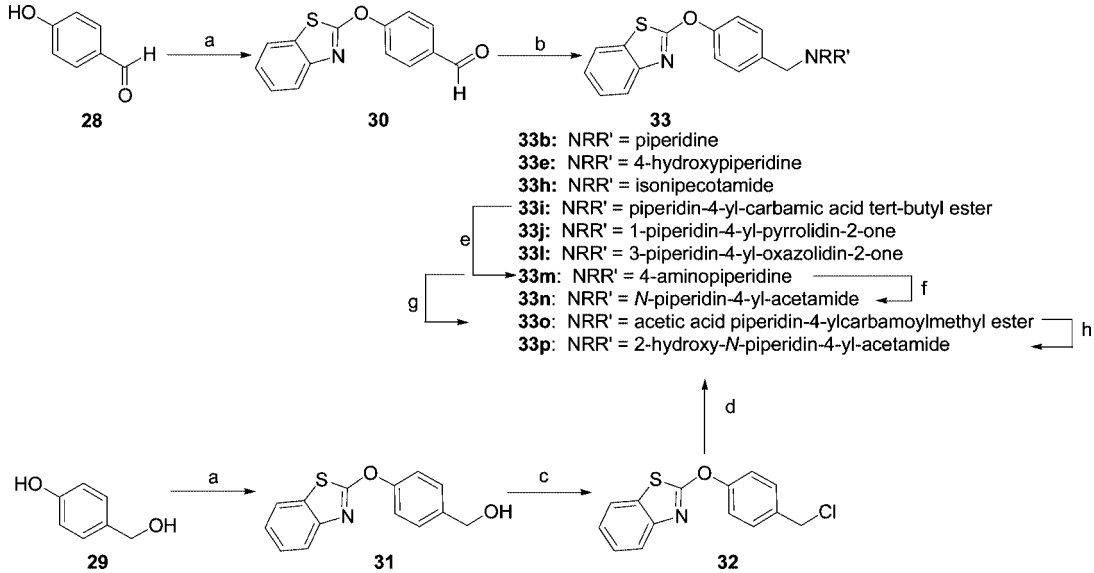
The one-carbon linker analogues were prepared using one of two methods as shown in Scheme 6. Treatment of 4-hydroxybenzaldehyde, **28**, with 2-chlorobenzothiazole in the presence of K_2CO_3 or Cs_2CO_3 followed by reductive amination using $NaBH(OAc)_3$ and various amines gave target compounds **33b**, **33e**, **33i**, and **33l** in yields ranging from 40% to 80%.[65] Alternatively, treatment of 4-hydroxybenzyl alcohol, **29**, with the 2-chlorobenzothiazole followed by treatment with thionyl chloride led to the formation of the chloride intermediate **32** in 50% overall yield. Formation of the target compounds **33h** and **33l** was accomplished using the desired amine in the presence of either Cs_2CO_3 or K_2CO_3. Select compounds are shown in Tables 4 and 5.[55]

Scheme 5. Synthesis of Analogues Containing a Two-Carbon Linker[a]



[a] (a) 2-Chlorobenzothiazole, Cs_2CO_3, CH_3CN, Δ; (b) 48% HBr, Δ; (c) PBr_3, benzene, Δ; (d) RR′NH, $^iPr_2EtN$; CH_3CN, Δ; (e) Dibal-H, toluene, −90 °C → −68 °C → −20 °C. (f) Preparation of **27h** and **27j**: RR′NH, silica-bound dimethylamine, CH_3CN, Δ. (g) Preparation of **27b** and **27e**: 2-chlorobenzothiazole, Cs_2CO_3, DMF, Δ. (h) Preparation of **27i** and **27l**: $NaBH(OAc)_3$, RR′NH, $ClCH_2CH_2Cl$, CH_3OH, room temp. (i) 88% HCO_2H, 5N HCl, room temp; (j) CH_3COCl, Et_3N, CH_2Cl_2, room temp; (k) acetoxyacetyl chloride, Et_3N, $ClCH_2CH_2Cl$, room temp.

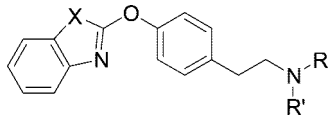
Scheme 6. Synthesis of Analogues Containing a One-Carbon Linker[a]



[a] (a) 2-Chlorobenzothiazole, K_2CO_3 or Cs_2CO_3, CH_3CN, Δ. (b) Preparation of **33b**, **33e**, **33i**, and **33j**: RR′NH, $NaBH(OAc)_3$, Et_3N, 4 Å molecular sieves, $ClCH_2CH_2Cl$; (c) $SOCl_2$, Et_3N, CH_2Cl_2, 5 °C → room temp. (d) Preparation of **33h** and **33l**: RR′NH, K_2CO_3 or Cs_2CO_3, CH_3CN, Δ; (e) 4 N HCl in dioxane, CH_2Cl_2, 0 °C → room temp; (f) acetyl chloride, Et_3N, CH_2Cl_2, rt; (g) acetoxyacetyl chloride, Et_3N, $ClCH_2CH_2Cl$, room temp; (h) LiOH, CH_2Cl_2, room temp.

The data in Tables 4 and 5 include enzymatic binding, inhibition of LTB_4 production in stimulated mouse whole blood (both in nM), and inhibition of astemizole binding to the hERG channel (in μM). Interestingly, manual docking studies suggested that the increased distance between the basic nitrogen in the one-carbon linker series analogues and the key acid residues Glu-271 and Glu-296 would result in a diminished interaction and that the potency of the one-carbon linker analogues would be attenuated. However, upon further inspection the neighboring Tyr-267 is appropriately positioned to allow for a π−cation interaction. Additionally, comparison of the inhibition of astemizole binding for the two-carbon and the one-carbon linker analogues reveals a significant reduction in hERG binding affinity for the one-carbon linker series.[66] Exceptions to this trend are noted, including a 2-fold decrease in the hERG binding for the pair of analogues **27n** and **33n**. From Table 5 above,

Table 4. In Vitro IC_{50} Data for Benzthiazole Compounds Containing the Two-Carbon Linker[54,a]



Compound Number	Amine NRR'	LTA_4H (nM)	MWB LTA_4H (nM)	HERG AB (μM)
27b		17 ± 6	280 ± 370	2
27e		8 ± 5	59 ± 41	3
27h		28 ± 25	300 ± 190	ND
27j		13 ± 11*	160 ± 50	1
27l		29 ± 29	160 ± 14	0.5
27n		35 ± 24	230 ± 52	9
27o		50 ± 42	510 ±270	5

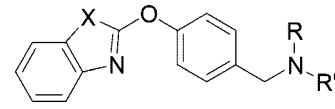
[a] Data expressed as IC_{50} ± std dev in nM. Murine whole blood diluted 1:14 with media is stimulated with calcium ionophore, A23187, and then assayed for LTB_4 production. (*) Average of two determinations.

we conclude that shortening of the linker length results in an attenuation of the hERG channel activity that may be due to decreased flexibility which minimizes favorable interactions with the ion channel. Several compounds in Tables 4 and 5 display favorable in vitro activity, thus warranting further profiling. The in vivo activity results of select compounds in the murine arachidonic acid induced ear inflammation model are provided in Table 6; the data for **10b** are included for comparative purposes. Compounds selected for evaluation in this assay would, in general, possess IC_{50} values in the enzymatic assay of <50 nM and IC_{50} values in the mouse whole blood assay of <300 nM. Preferred compounds would possess little to no activity in the hERG AB assay (>10 μM). Activity in this model is dependent on the pharmacokinetic properties of the compound in mouse. On the occasion that plasma concentrations at the single time point of $t = 4$ h postdose were determined, they were at or above the mouse whole blood IC_{50} value. This assay was used to screen compounds for advancement into rat or mouse pharmacokinetic studies. Despite the contrast of the data for compounds **27j** and **33j**, a clear preference for activity in this model with respect to linker length was not evident within a greater aggregate of results.

Advanced Profiling and In Vivo Evaluation

Compounds **33p** and **33r** afforded suitable in vitro and in vivo profiles as detailed above and were selected for additional evaluation. The syntheses of these two molecules are detailed in Schemes 6 and 7. For **33r**, Scheme 7, starting with commercially available **34**, treatment with the ethyl ester of isonipecotic acid resulted in the formation of **35** in 85% yield.

Table 5. In Vitro IC_{50} Data for Benzthiazole Compounds Containing the One-Carbon Linker[54,a]



Compound Number	Amine NRR'	LTA_4H (nM)	MWB LTA_4H (nM)	hERG AB (μM)
33b		59 ± 15	230 ± 150	>30
33e		34 ± 12	250 ± 250	>30
33h		17 ± 11	150 ± 160	>30
33j		12 ± 3	140 ± 36	15
33l		10 ± 13	110 ± 86	24
33n		12 ± 6	94 ± 62	20
33o		21 ± 13	210 ± 120	30
33p		8 ± 2	104 ± 100	>16
33r		11 ± 8	88 ± 24	>30

[a] Data expressed as IC_{50} ± std dev in nM. Murine whole blood diluted 1:14 with media is stimulated with calcium ionophore, A23187, and then assayed for LTB_4 production.

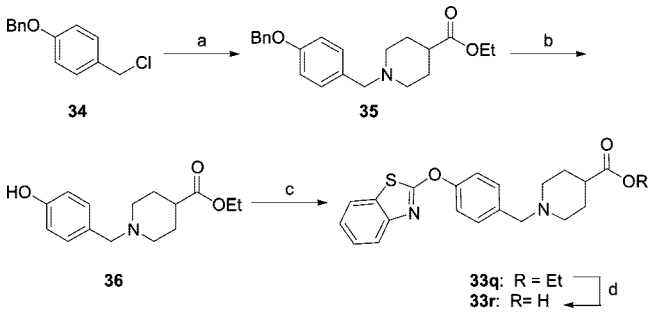
Table 6. In Vivo Data for Representative Analogues in the Murine Arachidonic Acid-Induced Ear Inflammation Model

compd	% inhibition of LTB_4 production	% inhibition of MPO activity
10b	77	77
11e	32	29
11f	79	83
11h	79	79
11j	67	84
27n	80	57
27j	51	44
33j	86	73
33l	95	74
33p	94	75
33q	81	82

Removal of the benzyl group was accomplished using 10% Pd/C in a 1:1 mixture of EtOAc and EtOH. Incorporation of the benzothiazole using standard conditions followed by saponification led to the formation of target compound **33r**.

While investigations of **33r** were recently reported,[57,67] we now disclose the biological data of **33p** (JNJ 27265732)[57] (Figure 8). Compound **33p** shows good inhibitory activity against the purified human enzyme and in diluted murine whole blood (IC_{50} = 7 and 84 nM, respectively). Reduction of the dilution ratio from 1:14 to 1:1 leads to an increase in the MWB LTA_4H IC_{50} to 425 nM, suggesting some binding to murine plasma proteins. This interaction was confirmed by plasma

Scheme 7. Alternative Synthesis of Compound **33r**[a]



[a] (a) Isonipecotic acid ethyl ester, K_2CO_3, CH_3CN, Δ; (b) 10% Pd/C, H_2, 40 psi, 1:1 EtOH/EtOAc; (c) 2-chlorobenzothiazole, K_2CO_3, CH_3CN, Δ; (d) KOH, 25% ^{i}PrOH/H_2O, room temp.

protein binding studies: **33p** shows moderately high binding to human (95%), murine (91%), rat (86%), and dog (84%) plasma proteins.[57] Compound **33p** was metabolically stable ($t_{1/2}$ > 60 min) when incubated in vitro with human, murine, rat, and dog microsomes or S9 fractions. This compound did not show any significant activity against a panel of 50 receptors and ion channels,[60] up to a concentration of 10 μM, nor any significant inhibition of the five major isoforms of human cytochrome P_{450} (IC_{50} > 10 μM). Compound **33p** was negative for mutagenicity in the Ames test.

In the murine model of arachidonic acid induced ear inflammation, **33p** dose-dependently blocked ex vivo production of LTB_4 in the blood when administered orally prior to inflammatory challenge with an estimated ED_{50} value for inhibition of LTB_4 production ex vivo of 3 mg/kg. This corresponds to an EC_{50} of ∼760 nM, which is similar to the in vitro murine blood IC_{50} (425 nM), determined under similar conditions. Similarly, significant inhibition of the neutrophil infiltration in response to topical arachidonic acid application to the ear was also seen; the ED_{50} was between 10 and 30 mg/kg (2 < EC_{50} < 6 μM). Further, comparative profiling of **33p**, **33r**, and the benzoxazole analogue **10b** was undertaken in a rat model of colitis induced by TNBS instillation, depicted in Figure 9.[68] Instillation of TNBS into vehicle treated animals resulted in a severe colitic inflammation after 3 days as measured by macroscopic lesion area and tissue LTB_4 and MPO content. Treatment with **33p**, **33r**, and **10b**, 30 mg/kg b.i.d., resulted in a significant reduction in all parameters.[69] Inhibition of lesion area was similar across compounds, with 42%, 49%, and 57% reduction, relative to vehicle treated and basal levels from nondiseased animals, for the respective compounds. For MPO, respective reductions of 81%, 72% and 63% were detected, while both **33r** and **33p** reduced LTB_4 content to basal levels.[70] The pharmacokinetic profiles of **33p** in both the rat and dog are shown in Figure 10 (**10b**, vide supra; **33r**, see ref 37). Compound **33p** has good bioavailability (65% and >100%, respectively) and intravenous half-life (3.5 and 3.6 h, respectively) in rats and dogs. The PK data in the dog suggest that the entire oral dose is absorbed with minimal loss to first pass metabolism.

Compound **33p** showed minimal inhibition of astemizole binding to the hERG channel (IC_{50} value of 16 μM); however, it showed 74% inhibition of the hERG-mediated K^+ current in a voltage clamp assay at 3 μM. Evaluation in the isolated guinea pig right atrium and the guinea pig ECG assays did not show any significant compound-related effects up to 10 and 21 μM, respectively. Given the encouraging data that have been accumulated, compound **33p** is a viable candidate for further preclinical evaluation.

Conclusion

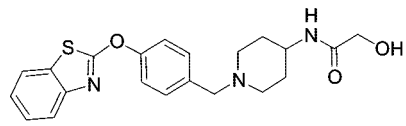
We have described the identification and structural optimization of benzthiazole, benzoxazole, and benzimidazole LTA_4H inhibitors. Our work has focused primarily on modifications to the linker length and the basic amine. Novel benzoxazole and benzthiazole analogues have been prepared that inhibit the function of LTA_4H in vivo when dosed orally. The poor stability at low pH of the benzoxazole analogues, although not profoundly affecting the in vivo efficacy, would clearly complicate long-term developability and as such further studies focused on the benzthiazole series of analogues. Additionally, extensive SAR studies indicated that a good compromise between hERG binding affinity (as measured by inhibition of astemizole binding) and in vivo efficacy in the murine arachidonic acid induced ear inflammation model could be attained and resulted in the identification of **33p**. Further optimization of these inhibitors will be the subject of additional disclosures from our laboratories.

Experimental Section

General. Except where indicated, materials and reagents were used as supplied from commercial vendors. Reaction monitoring was performed with EMD silica gel 60 F254 250 μm precoated TLC plates and visualized with UV light. Routine chromatographic purifications were performed via semiautomated MPLC, using prepacked RediSep 35−60 μm silica gel columns on ISCO Sg100 systems with UV peak detection. Analytical LC spectra were collected on an Agilent/HP1100 LC system with diode array UV (230, 254, and 280 nm) detection and a MeCN/water/0.05% TFA solvent gradient running on a Phenomenex Luna C18(2) 5 μm column. MS data were recorded on an Agilent/HP100 LC system coupled to an Agilent single quadrupole electrospray MSD. Proton NMR spectra were recorded at either 400 or 500 MHz using Bruker DPX-400 and DPX-500 spectrometers and were taken in $CDCl_3$ at ambient temperature unless otherwise noted. Coupling constants are expressed in Hz. High-resolution mass spectra (HRMS) were determined on a Bruker micro-TOF instrument with internal calibration based on sodium formate ion clusters. Combustion analyses were performed by NuMega Resonance Laboratories, Inc., San Diego, CA. Bioanalytical quantitation was performed via mass spectrometry using either a Finnigan Quantum Ultra system (Thermo Electron Corp.) or an API-4000 system (Applied Biosystems).

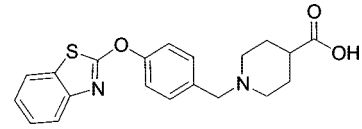
2-(4-Benzyloxyphenoxy)ethyl Bromide (6). To a stirring solution of 4-benzyloxyphenol (72 g, 360 mmol) in CH_3CN (600 mL) was added dibromoethane (155 mL, 1.80 mol) and K_2CO_3 (105 g, 760 mmol). This brown suspension was heated at reflux and allowed to stir for 96 h. The resulting suspension was cooled to room temperature, diluted with acetone (250 mL), and filtered through diatomaceous earth, which was then rinsed with additional acetone. The filtrate was concentrated under reduced pressure. The resulting oil was dissolved in CH_3OH (500 mL), and the solution was stirred for 2 h. The title compound was obtained by filtration and air-dried to give 70 g (230 mmol, 63% yield) as a tan solid. ^{1}H NMR (400 MHz, $CDCl_3$): 7.60−7.30 (m, 5H), 6.88 (d, J = 8.4, 2H), 6.80 (d, J = 8.4, 2H), 4.70 (s, 2H), 3.79 (t, J = 5.8, 2H), 3.07 (t, J = 5.8, 2H).

4-(2-Bromoethoxy)phenol (7). Compound **6** (70 g, 230 mmol) was dissolved in THF (500 mL). To this solution was added Pd on carbon (10 wt %, 7 g) as a suspension in ethanol (50 mL). The resulting suspension was placed on a Parr hydrogenator at 40 psi of H_2 and shaken overnight. The reaction mixture was filtered through a pad of diatomaceous earth, and the filtrate was concentrated under reduced pressure to give 48.5 g (224 mmol, 99% yield) of a tan solid. ^{1}H NMR (400 MHz, $CDCl_3$): 6.83 (d, J = 9.1, 2H),



33p

***In Vitro* Data**
LTA$_4$H IC$_{50}$ = 8 ± 2 nM
MWB LTA$_4$H IC$_{50}$ =104 ± 100 nM
Cardiovascular:
hERG AB IC$_{50}$ = 16 μM
***In Vivo* Data; 30 mg/Kg, po**
% Inh LTB$_4$ = 94%
% Inh MPO = 75%



33r

***In Vitro* Data**
LTA$_4$H IC$_{50}$ = 11 ± 8 nM
MWB LTA$_4$H IC$_{50}$ = 88 ± 24 nM
Cardiovascular:
hERG AB IC$_{50}$ > 30 μM
***In Vivo* Data; 30 mg/Kg, po**
% Inh LTB$_4$ = 80%
% Inh MPO = 94%

Figure 8. In vitro and in vivo data for **33p** and **33r**.

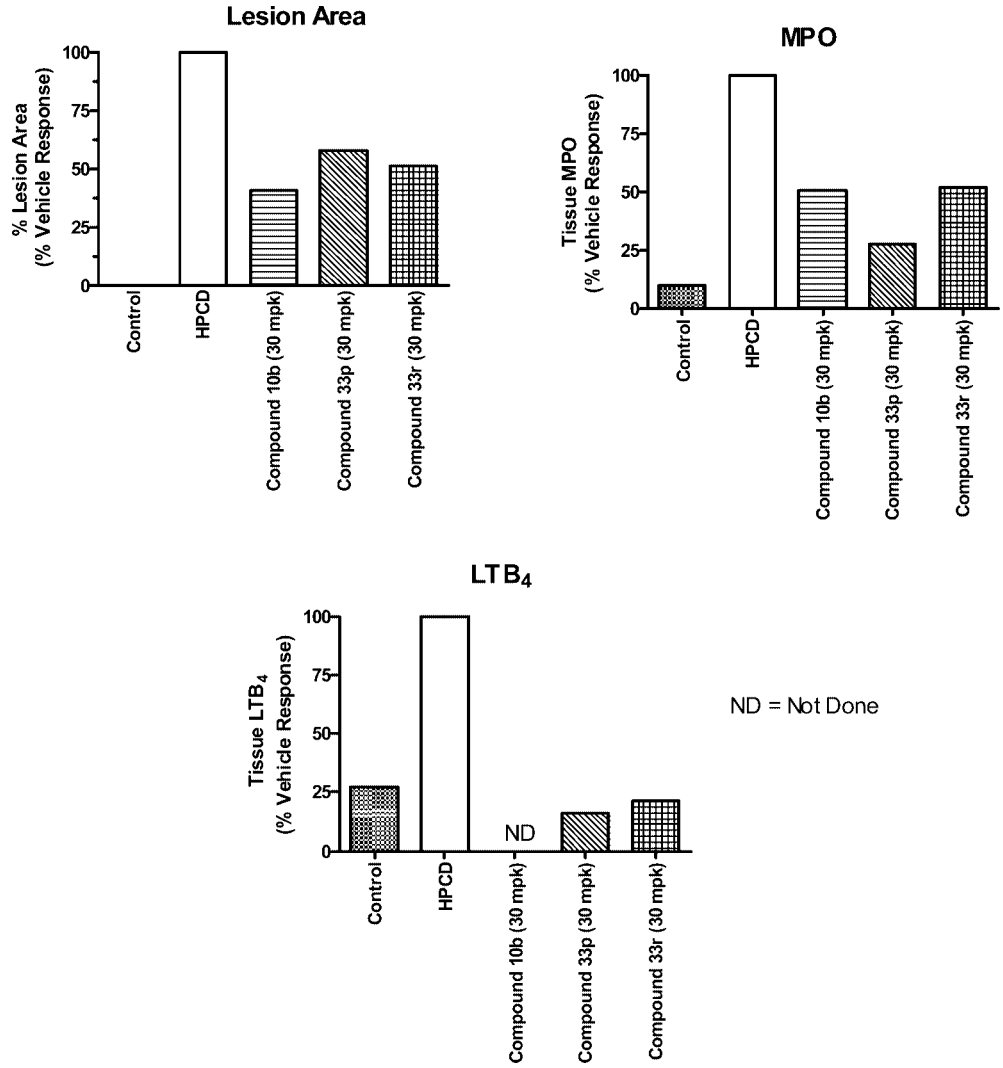
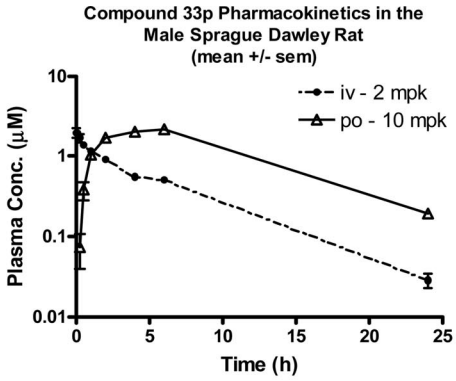


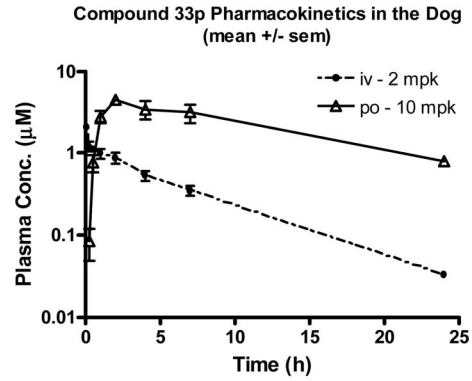
Figure 9. Comparison of **10b**, **33p**, and **33r** in the TNBS model of colitis in the rat.[66]

6.77 (d, J = 9.1, 2H), 4.51 (s, 1H), 4.24 (t, J = 6.3, 2H), 3.62 (t, J = 6.3, 2H).

1-[2-(4-Hydroxyphenoxy)ethyl]piperidin-4-ol (9e). To a stirring solution of **7** (9.0 g, 42 mmol) in CH_3CN (150 mL) was added 4-hydroxypiperidine (5.3 g, 53 mmol), followed by *N*,*N*-diisopropylethylamine (6.7 g, 53 mmol). The resulting solution was stirred overnight at room temperature, yielding a suspension. The suspension was filtered, and the filtrate was concentrated under reduced pressure. Diethyl ether was added to the resultant oil, and the mixture was warmed to 45 °C for 2 min, forming a white precipitate. This suspension was stirred at room temperature for 2 h, then filtered, giving 7.9 g (33 mmol, 79% yield) of an off-white solid. MS (ESI): mass calculated for $C_{13}H_{19}NO_3$, 237.14; *m*/*z* found, 238.2 $[M + H]^+$. ^{1}H NMR (400 MHz, CD_3OD): 7.10 and 7.02 (q, J = 32.3, 9.0, 2H), 4.35 (t, J = 5.7, 2H), 3.59 (m, 1H), 3.26−3.19 (m, 2H), 3.07 (t, J = 5.7, 2H), 2.60 (t, J = 9.9, 2H), 2.20−2.12 (m, 2H), 1.94−1.82 (m, 2H).

1-[2-(4-Benzyloxy-phenoxy)ethyl]piperidine (8b). To a mixture of 4-benzyloxyphenol (24.6 g, 123 mmol) and 1-(2-chloroethyl)piperidine hydrochloride (20.6 g, 112 mmol) in DMF (175 mL) was





Route	Dose (mg/Kg)	t_{max} (h)	C_{max} (μmol/L)	AUC (h*μmol/L)	CL (L/h)	V_{ss} (L/Kg)	$t_{½}$ (h)	%F
Rat iv	2	--	2.2	10	0.18	2.5	3.5	--
Rat po	10	5.3	2.2	33	--	--	5.6[a]	65
Dog iv	2	--	2.8	8.0	5.6	3.2	3.6	--
Dog po	10	1.0	26	60.3	--	--	6.9	150

[a]Half-life is an estimate of the terminal elimination phase from the oral dose.

Figure 10. Pharmacokinetic profile of **33p** in the rat and dog.

added K_2CO_3 (25.0 g, 181 mmol) and Cs_2CO_3 (40.0 g, 123 mmol). The reaction mixture was stirred for 3 days at room temperature. To the mixture was added H_2O (300 mL) and CH_2Cl_2. The organic layer was washed with 10% NaOH and then brine, dried ($MgSO_4$), filtered, and concentrated under reduced pressure to give 33 g of a clear, dark-purple liquid. The liquid was purified on SiO_2 (300 g, 0−50% ethyl acetate/hexanes) to give 23.4 g (75 mmol, 67% yield) of a light-yellow solid. TLC (SiO_2, 50% hexanes/ethyl acetate): R_f = 0.11. MS (ESI): mass calculated for $C_{20}H_{25}NO_2$, 311.19; *m/z* found, 312.2 $[M + H]^+$. ^1H NMR (400 MHz, $CDCl_3$): 7.50−7.26 (m, 5H), 6.91 (d, J = 9.2, 2H,), 6.85 (d, J = 9.2, 2H), 5.02 (s, 2H), 4.06 (t, J = 6.1, 2H), 2.76 (t, J = 6.1, 2H), 2.51 (br s, 4H), 1.65−1.55 (m, 4H), 1.45 (br s, 2H).

4-(2-Piperidin-1-ylethoxy)phenol (9b). To a solution of **8b** (15.0 g, 48.2 mmol) in 1:1 ethanol/ethyl acetate (400 mL) was added Pd on carbon (10 wt %, 1.5 g). The mixture was placed on a Parr hydrogenator at 40 psi of H_2 for 20 h. The reaction mixture was filtered through diatomaceous earth, and the filtrate was concentrated under reduced pressure to give 9.4 g (43.5 mmol, 88% yield) of the desired product as a light-gray solid. TLC (SiO_2, 50% acetone/CH_2Cl_2): R_f = 0.16. MS (ESI): mass calculated for $C_{13}H_{19}NO_2$, 221.14; *m/z* found, 222.1 $[M + H]^+$. ^1H NMR (400 MHz, DMSO-d_6): 8.88 (s, 1H), 6.73 (d, J = 6.6, 2H), 6.65 (d, J = 6.6, 2H), 3.93 (t, J = 6.0, 2H), 2.58 (t, J = 6.0, 2H), 2.40 (s, 4H), 1.51−1.45 (m, 4H), 1.35 (br s, 2H).

2-Chloro-1-(2-trimethylsilanylethoxymethyl)-1*H*-benzoimidazole (14). To a suspension of sodium hydride (6.2 g, 240 mmol) in DMF (275 mL) at 5 °C was added 2-chlorobenzimidazole (37.0 g, 240 mmol) via a solid-addition funnel over 30 min while maintaining the internal temperature of the mixture below 10 °C. An additional 25 mL of DMF was added, and the ice bath was removed. After 2 h, 2-(trimethylsilyl)ethoxymethyl chloride (SEM-Cl, 44.0 g, 264 mmol) was added dropwise over 5 min. A white precipitate formed. The reaction mixture was stirred at room temperature for 18 h. To the mixture was added H_2O (500 mL) and ethyl acetate (750 mL). The organic layer was washed with additional H_2O (500 mL), dried ($MgSO_4$), and concentrated under reduced pressure, giving 65.8 g (233 mmol, 96% yield) of the desired product as a clear golden oil, which solidified upon standing to give a beige solid. TLC (SiO_2, 5% acetone/CH_2Cl_2): R_f = 0.64. MS (ESI): mass calculated for $C_{13}H_{19}ClN_2OSi$, 282.10; *m/z* found, 283.1. ^1H NMR (400 MHz, $CDCl_3$): 7.70 (d, J = 7.3, 1H), 7.46 (d, J = 7.6, 1H), 7.40−7.25 (m, 2H), 3.58 (t, J = 7.9, 2H), 0.92 (t, J = 8.3, 2H), 0.04 (s, 9H).

2-[4-(2-Bromoethoxy)phenoxy]benzothiazole (15). A solution of **7** (8.7 g, 40 mmol) and 2-chlorobenzothiazole (12.0 mL, 92 mmol) in CH_3CN was treated with finely powdered Cs_2CO_3 (26.0 g, 80 mmol), and the resulting mixture was stirred at 23 °C for 30 h. The reaction mixture was filtered through diatomaceous earth, and the filtrate was concentrated under reduced pressure. The crude solid was purified on SiO_2 (100 g, 0−40% ethyl acetate/hexanes) to provide 6.7 g (19 mmol, 47% yield) of a white solid. MS (ESI): mass calculated for $C_{15}H_{12}BrNO_2S$, 348.98; *m/z* found, 350.0 $[M + H]^+$. ^1H NMR (400 MHz, $CDCl_3$): 7.78, (dd, J = 8.0, 0.4, 1H), 7.70 (dd, J = 8.0, 0.7, 1H), 7.42 (dt, J = 7.5, 1.3, 1H), 7.34 (dd, J = 9.1, 2H), 7.01 (dd, J = 9.1, 2H), 4.34 (t, J = 6.2, 2H), 3.70 (t, J = 6.2, 2H).

2-[4-(2-Pyrrolidin-1-ylethoxy)phenoxy]benzooxazole (10a). The title compound was prepared according to the procedures for **8b** and **10b** utilizing an analogous intermediate **8a** derived from 1-(2-chloroethyl)pyrrolidine to provide 1.9 g of **10a** as a beige solid (5.9 mmol, 90% yield). MS (ESI): mass calculated for $C_{19}H_{20}N_2O_3$, 324.15; *m/z* found, 325.1 $[M + H]^+$. ^1H NMR (400 MHz, $CDCl_3$): 7.75 (d, J = 8.1,1H), 7.66 (d, J = 8.0, 1H), 7.40 (dt, J = 7.4, 1.3, 1H), 7.31−7.23 (m, 3H), 6.98 (d, J = 6.8, 2H), 4.08 (t, J = 6.3, 2H), 2.91 (t, J = 6.3, 2H), 2.67 (q, J = 7.2, 4H), 1.10 (t, J = 7.2, 6H). Acc Mass ($C_{19}H_{20}N_2O_2$). Anal. ($C_{19}H_{20}N_2O_3$) C, H, N.

2-[4-(2-Pyrrolidin-1-ylethoxy)phenoxy]benzothiazole (11a). The title compound was prepared according to the procedure for **8b** and **11e** utilizing an analogous intermediate **8a** derived from 1-(2-chloroethyl)pyrrolidine and 2-chlorobenzothiazole to provide 785 mg of **11a** as a clear golden oil (23 mmol, 20% yield). MS (ESI): mass calculated for $C_{19}H_{20}N_2O_2S$, 340.12; *m/z* found, 341.1 $[M + H]^+$. ^1H NMR (400 MHz, $CDCl_3$): 7.77 (d, J = 7.6, 1H), 7.66 (d, J = 8.6, 1H), 7.41 (t, J = 7.4, 1H), 7.32−7.25 (m, 3H), 7.00 (d, J = 9.0, 2H), 4.16 (t, J = 6.0, 2H), 2.96 (t, J = 6.0, 2H), 2.67 (t, J = 6.6, 4H), 1.86 (quint, J = 3.5, 4H). Anal. ($C_{19}H_{20}N_2O_2S$) C, H, N.

2-[4-(2-Pyrrolidin-1-ylethoxy)phenoxy]-1*H*-benzoimidazole (13a). The title compound was prepared according to the procedures for **12d** and **13d**, using 4-(2-pyrrolidin-4-ylethoxy)phenol prepared according to the procedure for **8b** derived from 1-(2-chloroethyl)pyrrolidine to provide 21 mg of **13a** as a white solid (0.07 mmol, 19% yield). MS (ESI): mass calculated for $C_{19}H_{21}N_3O_2$, 323.16; *m/z* found, 324.2 $[M + H]^+$. ^{1}H NMR (400 MHz, $CDCl_3$): 7.27 (br s, 2H), 7.15 (d, $J = 9.1$, 4H), 6.81 (d, $J = 6.8$, 2H), 4.16 (t, $J = 5.5$, 2H), 3.05 (t, $J = 5.5$, 2H), 2.86 (br s, 4H), 1.92 (br s, 4H).

2-[4-(2-Piperidin-1-ylethoxy)phenoxy]benzooxazole (10b). To a stirred solution of **9b** (1.5 g, 6.8 mmol) in acetone (20 mL) at 5 °C was added K_2CO_3 (1.0 g, 7.2 mmol). To the mixture was added 2-chlorobenzoxazole (0.5 mL, 4.4 mmol) at 5 °C. The resulting mixture was warmed to room termperature overnight. After 20 h the mixture was filtered, and the filtrate was concentrated under reduced pressure to a brown solid, which was purified on SiO_2 (35 g, 50% acetone/CH_2Cl_2). The desired fractions were combined and concentrated under reduced pressure to give 1.2 g (3.5 mmol, 80% yield) of the desired product as a white solid. TLC (SiO_2, 50% acetone/CH_2Cl_2): $R_f = 0.18$. MS (ESI): mass calculated for $C_{20}H_{22}N_2O_3$, 338.16; *m/z* found, 339.1 $[M + H]^+$. ^{1}H NMR (400 MHz, $CDCl_3$): 7.52 (d, $J = 7.2$, 1H), 7.41 (d, $J = 7.2$, 1H), 7.35−7.20 (m, 4H), 6.97 (d, $J = 9.1$, 2H), 4.12 (t, $J = 6.1$, 2H), 2.79 (t, $J = 6.0$, 2H), 2.52 (s, 4H), 1.67−1.55 (m, 4H), 1.50−1.40 (m, 2H). Anal. ($C_{20}H_{22}N_2O_3$) C, H, N.

2-[4-(2-Piperidin-1-ylethoxy)phenoxy]benzothiazole (11b). The title compound was prepared according to the procedure for **8b** and **11e** to provide 450 mg of **11b** as a clear orange oil (1.3 mmol, 56% yield). MS (ESI): mass calculated for $C_{20}H_{22}N_2O_2S$, 354.14; *m/z* found, 355.1 $[M + H]^+$. ^{1}H NMR (400 MHz, $CDCl_3$): 7.74 (d, $J = 8.1$, 1H), 7.65 (d, $J = 8.0$, 1H), 7.39 (t, $J = 7.3$, 1H), 7.30−7.22 (m, 3H), 7.00−6.92 (m, 2H), 4.13 (t, $J = 6.0$, 2H), 2.81 (t, $J = 6.0$, 2H), 2.58−2.48 (br s, 4H), 1.68−1.58 (m, 4H), 1.52−1.42 (m, 2H). Anal. ($C_{20}H_{22}N_2O_2S$) C, H, N.

2-[4-(2-Piperidin-1-ylethoxy)phenoxy]-1*H*-benzoimidazole (13b). The title compound was prepared according to the procedure for **12d** and **13d**, using 4-(2-piperidin-4-ylethoxy)phenol prepared according to the procedure for **8b** using 1-(2-chloroethyl)piperidine to provide 1.2 g of **13b** as a white solid (3.6 mmol, 75% yield). MS (ESI): mass calculated for $C_{20}H_{23}N_3O_2$, 337.18; *m/z* found, 338.3 $[M + H]^+$. ^{1}H NMR (400 MHz, DMSO-d_6): 12.25 (br s, 1H), 7.35−7.25 (m, 4H), 7.10−7.00 (m, 2H), 6.99 (d, $J = 9.0$, 2H), 4.07 (t, $J = 5.9$, 2H), 2.79 (t, $J = 5.9$, 2H), 2.43 (s, 4H), 1.55−1.45 (m, 4H), 1.38 (s, 2H). Anal. ($C_{19}H_{21}N_3O_3 \cdot 0.25H_2O$) C, H, N.

2-[4-(2-Azepan-1-ylethoxy)phenoxy]benzooxazole (10c). The title compound was prepared according to the procedure for **8b** and **10b** utilizing an analogous intermediate **8c** derived from 1-(2-chloroethyl)azepane hydrochloride 1.2 g of **10c** as a pale-yellow oil (3.4 mmol, 88% yield). MS (ESI): mass calculated for $C_{21}H_{24}N_2O_3$, 352.18; *m/z* found, 353.2 $[M + H]^+$. ^{1}H NMR (400 MHz, $CDCl_3$): 7.49−7.47 (m, 1H), 7.41−7.38 (m, 1H), 7.31−7.27 (m, 2H), 7.26−7.18 (m, 2H), 6.97−6.93 (m, 2H), 4.06 (t, $J = 6.2$, 2H), 2.94 (t, $J = 6.2$, 2H), 2.77−2.75 (m, 4H), 1.65−1.58 (m, 8H). Anal. ($C_{21}H_{24}N_2O_3$) C, H, N.

2-[4-(2-Azepan-1-ylethoxy)phenoxy]benzothiazole (11c). The title compound was prepared according to the procedure for **8b** and **11e** utilizing an analogous intermediate **8c** derived from 1-(2-chloroethyl)azepane hydrochloride and 2-chlorobenzothiazole 375 mg of **11c** as an off-white solid (0.93 mmol, 48% yield). MS (ESI): mass calculated for $C_{21}H_{24}N_2O_2S$, 368.16; *m/z* found, 369.2 $[M + H]^+$. ^{1}H NMR (400 MHz, $CDCl_3$): 7.70 (d, $J = 7.5$, 1H), 7.65 (d, $J = 7.9$, 1H), 7.38−7.34 (m, 1H), 7.29−7.23 (m, 3H), 6.97 (br d, 2H), 4.59 (br s, 2H), 3.64 (br s, 2H), 3.46 (br s, 2H), 3.13 (br s, 3H), 2.19 (br s, 2H), 1.87 (br s, 2H), 1.72−1.58 (m, 2H). Anal. ($C_{21}H_{24}N_2O_2S \cdot HCl$) C, H, N.

2-[4-(2-Azepan-1-ylethoxy)phenoxy]-1*H*-benzoimidazole amide (13c). The title compound was prepared according to the procedure for **12d** and **13d**, using 4-(2-azepan-4-ylethoxy)phenol prepared according to the procedure for **8c** using 1-(2-chloro-ethyl)azepane to provide 142 mg of **13c** as a white solid (0.4 mmol, 35% yield). MS (ESI): mass calculated for $C_{21}H_{25}N_3O_2$, 351.19; *m/z* found, 352.3 $[M + H]^+$. ^{1}H NMR (400 MHz, DMSO-d_6): 12.28 (s, 1H), 7.35−7.25 (m, 4H), 7.07 (d, $J = 9.1$, 2H), 7.00 (d, $J = 9.1$, 2H), 4.05 (t, $J = 6.0$, 2H), 2.86 (t, $J = 6.0$, 2H), 2.70 (t, $J = 5.1$, 4H), 1.65−1.50 (m, 8H). Anal. ($C_{21}H_{25}N_3O_2$) C, H, N.

2-[4-(2-Morpholin-4-ylethoxy)phenoxy]benzooxazole (10d). The title compound was prepared according to the procedure for **8b** and **10b** utilizing an analogous intermediate **8d** derived from 4-(2-chloroethyl)morpholine to provide 197 mg of **10d** as a white solid (0.6 mmol, 52% yield). MS (ESI): mass calculated for $C_{19}H_{20}N_2O_4$, 340.14; *m/z* found, 341.1 $[M + H]^+$. ^{1}H NMR (400 MHz, $CDCl_3$): 7.51 (d, $J = 7.1$, 1H), 7.42 (d, $J = 7.2$, 1H), 7.36−7.30 (m, 2H), 7.29−7.20 (m, 2H), 7.03−6.95 (m, 2H), 4.14 (t, $J = 5.7$, 2H), 3.80−3.72 (m, 4H), 2.83 (t, $J = 5.7$, 2H), 2.60 (t, $J = 4.6$, 4H). Anal. ($C_{19}H_{20}N_2O_4$) C, H, N.

2-[4-(2-Morpholin-4-ylethoxy)phenoxy]benzothiazole (11d). The title compound was prepared according to the procedure for **8b** and **11e** utilizing an analogous intermediate **8d** derived from 4-(2-chloroethyl)morpholine and 2-chlorobenzothiazole to provide 231 mg of **11d** as a clear golden oil (0.7 mmol, 58% yield). MS (ESI): mass calculated for $C_{19}H_{20}N_2O_3S$, 35; *m/z* found, 341.1 $[M + H]^+$. ^{1}H NMR (400 MHz, $CDCl_3$): 7.74 (d, $J = 7.6$, 1H), 7.66 (d, $J = 7.3$, 1H), 7.39 (t, $J = 7.3$, 1H), 7.31−7.20 (m, 3H), 7.00−6.93 (m, 2H), 4.14 (t, $J = 5.7$, 2H), 3.80−3.72 (m, 4H), 2.83 (t, $J = 5.7$, 2H), 2.60 (t, $J = 4.6$, 4H). Anal. ($C_{19}H_{20}N_2O_3S$) C, H, N.

2-[4-(2-Morpholin-4-ylethoxy)phenoxy]-1-(2-trimethylsilanylethoxymethyl)-1*H*-benzoimidazole (12d). To a mixture of **14** (570 mg, 2.0 mmol) and **9d** (prepared according to the procedure for **9b** using 4-(2-chloroethyl)morpholine, 450 mg, 2.0 mmol) in DMF (10 mL) was added Cs_2CO_3 (1.4 g, 4.3 mmol). The reaction mixture was stirred at 100 °C for 18 h and then partitioned in 1:1 ethyl acetate/H_2O (50 mL). The organic layer was collected, dried ($MgSO_4$), and concentrated under reduced pressure to give a clear brown oil, which was purified on SiO_2 (35 g, 50% acetone/CH_2Cl_2) to give 730 mg (1.6 mmol, 78% yield) of the desired product as a clear golden oil. TLC (SiO_2, acetone): $R_f = 0.69$. MS (ESI): mass calculated for $C_{25}H_{35}N_3O_4Si$, 469.24; *m/z* found, 470.2 $[M + H]^+$. ^{1}H NMR (400 MHz, $CDCl_3$): 7.58−7.54 (m, 1H), 7.40−7.37 (m, 1H), 7.27 (d, $J = 4.54$ Hz, 2H), 7.23−7.19 (m, 2H), 6.97 (d, $J = 9.09$ Hz, 2H), 5.55 (s, 2H), 4.13 (t, $J = 5.69$ Hz, 2H), 3.78−3.73 (m, 4H), 3.70−3.64 (m, 2H), 2.82 (t, $J = 5.69$ Hz, 2H), 2.62−2.56 (m, 4H), 1.00−0.91 (m, 2H), 0.00 (s, 9H).

2-[4-(2-Morpholin-4-ylethoxy)phenoxy]-1*H*-benzoimidazole (13d). To a solution of **12d** (675 mg, 1.4 mmol) in THF (5 mL) containing *N*,*N*,*N*,*N*-tetramethylethylenediamine (TMEDA, 2.2 mL, 14.5 mmol) was added a 1 M THF solution of tetrabutylammonium fluoride (TBAF, 15 mL, 15.0 mmol). The mixture was stirred at 55 °C for 5 h and was concentrated under reduced pressure. The resulting oil was dissolved in diethyl ether (100 mL), and the solution was washed with H_2O (3 × 75 mL), dried ($MgSO_4$), filtered, and concentrated under reduced pressure to give a white solid. Diethyl ether was added, and filtration gave 297 mg of the desired product as a white solid (0.9 mmol, 61% yield). TLC (SiO_2, acetone): $R_f =$ 0.58. MS (ESI): mass calculated for $C_{19}H_{21}N_3O_3$, 339.16; *m/z* found, 340.2 $[M + H]^+$. ^{1}H NMR (400 MHz, DMSO-d_6): 7.40−7.25 (m, 4H), 7.12−7.08 (m, 2H), 7.00 (d, $J = 9.0$, 2H), 4.10 (t, $J = 5.7$, 2H), 3.58 (t, $J = 4.5$, 4H), 2.70 (t, $J = 5.7$, 2H), 2.52−2.46 (m, 4H). Anal. ($C_{19}H_{21}N_3O_3$) C, H, N.

1-{2-[4-(Benzooxazol-2-yloxy)phenoxy]ethyl}piperidin-4-ol (10e). The title compound was prepared in an analogous manner to **10b** using 1-[2-(4-hydroxyphenoxy)ethyl]piperidin-4-ol and K_2CO_3 to provide 910 mg of **10e** as a white solid (2.6 mmol, 61% yield). MS (ESI): mass calculated for $C_{20}H_{22}N_2O_4$, 354.16; *m/z* found, 355.2 $[M + H]^+$. ^{1}H NMR: (400 MHz, $CDCl_3$) 7.55 (dd, $J = 7.2$, 1.8, 1H), 7.46 (dd, $J = 7.3$, 2.0, 1H), 7.36 (d, $J = 9.1$, 2H), 7.32−7.25 (m, 2H), 7.01 (d, $J = 9.1$, 2H), 4.18 (t, $J = 5.4$, 2H), 3.80 (m, 1H), 3.01−2.86 (m, 4H), 2.40 (br s, 1H), 1.99 (m, 2H), 1.74−1.65 (m, 2H), 1.48 (d, $J = 4.1$, 1H). Anal. ($C_{20}H_{22}N_2O_4$) C, H, N.

1-{2-[4-(Benzothiazol-2-yloxy)phenoxy]ethyl}piperidin-4-ol (11e). To a stirring solution of 1-[2-(4-hydroxyphenoxy)ethyl]piperidin-4-ol (500 mg, 1.3 mmol) in DMF (10 mL) was added Cs_2CO_3 (1.4 g, 4.4 mmol) and 2-chlorobenzothiazole (0.33 mL, 2.5 mmol). The suspension was heated to 80 °C and stirred overnight. The reaction mixture was allowed to cool to room temperature and then filtered through diatomaceous earth. The filtrate was concentrated under reduced pressure, and the residue was purified on SiO_2 (40 g, 0−100% acetone/CH_2Cl_2), giving 321 mg (0.9 mmol, 69% yield) of a tan solid. MS (ESI): mass calculated for $C_{20}H_{22}N_2O_3S$, 370.14; *m/z* found, 371.2 $[M + H]^+$. ^1H NMR (400 MHz, $CDCl_3$): 7.64 (d, J = 8.0, 1H), 7.56 (d, J = 7.8, 1H), 7.30 (t, J = 7.2, 1H), 7.21−7.14 (m, 3H), 6.87 (d, J = 9.1, 2H), 4.05 (t, J = 5.8, 2H), 3.67 (br s, 1H), 2.82 (m, 2H), 2.76 (t, J = 5.8, 2H), 2.27 (t, J = 9.5, 2H), 2.01 (br s, 1H), 1.90−1.82 (m, 2H), 1.64−1.52 (m, 2H). Anal. ($C_{20}H_{22}N_2O_3S \cdot 0.3H_2O$) C, H, N.

(1-{2-[4-Benzooxazol-2-loxy)phenoxy]ethyl}piperidin-4-yl)methanol (10f). The title compound was prepared according to the procedure for **10b** using **9f** to provide 424 mg of **10f** as a white solid (1.2 mmol, 58% yield). TLC (SiO_2, 5% 2 M NH_3 in CH_3OH/CH_2Cl_2): R_f = 0.17. MS (ESI): mass calculated for $C_{21}H_{24}N_2O_4$, 368.17; *m/z* found, 369.3 $[M + H]^+$. ^1H NMR (400 MHz, $CDCl_3$): 7.53−7.50 (m, 1H), 7.44−7.42 (m, 1H), 7.34−7.21 (m, 4H), 7.00−6.96 (m, 2H), 4.13 (t, J = 6.0, 2H,), 3.55−3.48 (m, 2H), 3.02−3.09 (m, 2H), 2.82 (t, J = 6.0, 2H), 2.13 (dt, J = 2.4, 11.8, 2H), 1.78−1.75 (m, 2H), 1.59−1.48 (m, 2H), 1.33 (dq, J = 3.7, 12.4, 2H). Anal. ($C_{21}H_{24}N_2O_4$) C, H, N.

(1-{2-[4-(Benzothiazol-2-yloxy)phenoxy]ethyl}piperidin-4-yl)-methanol (11f). The title compound was prepared according to the procedures for **9e** and **11e** using piperidin-4-ylmethanol to provide 275 mg of **11f** as a white solid (0.72 mmol, 45% yield). MS (ESI): mass calculated for $C_{21}H_{24}N_2O_3S$, 384.15; *m/z* found, 385.1 $[M + H]^+$. ^1H NMR (500 MHz, $CDCl_3$): 7.75−7.73 (m, 1H), 7.67−7.65 (m, 1H), 7.41−7.37 (m, 1H), 7.31−7.36 (m, 3H), 6.98−6.95 (m, 2H), 4.13 (t, J = 5.9, 2H), 3.52 (t, J = 5.4, 2H), 3.10−3.03 (m, 2H), 2.83 (t, J = 5.9, 2H), 2.14 (dt, J = 11.8, 2.4, 2H), 1.78−1.75 (m, 2H), 1.58−1.52 (m, 2H), 1.37−1.30 (m, 2H). Anal. ($C_{21}H_{24}N_2O_3S$) C, H, N.

1-{2-[4-(Benzooxazol-2-yloxy)phenoxy]ethyl}-4-phenylpiperidin-4-ol (10g). The title compound was prepared according to the procedure for **9e** and **10b** using 4-phenylpiperidin-4-ol to provide 600 mg of **10g** as a white solid (1.4 mmol, 87% yield). MS (ESI): mass calculated for $C_{26}H_{26}N_2O_4$, 430.19; *m/z* found, 431.2 $[M + H]^+$. ^1H NMR (400 MHz, $CDCl_3$): 7.60−7.45 (m, 3H), 7.40−7.18 (m, 8H), 7.00 (d, J = 9.1, 2H), 4.18 (t, J = 5.9, 2H), 2.92 (t, J = 5.9, 2H), 2.66 (dt, J = 12.1, 2.4, 2H), 2.22 (dt, J = 13.4, 4.5, 2H), 1.80 (dd, J = 14.1, 2.4, 2H). Anal. ($C_{26}H_{26}N_2O_4$) C, H, N.

1-{2-[4-(Benzothiazol-2-yloxy)phenoxy]ethyl}-4-phenylpiperidin-4-ol (11g). The title compound was prepared according to the procedure for **9e** and **11e** using 4-phenylpiperidin-4-ol to provide 570 mg of **11g** as a light-yellow solid (1.3 mmol, 80% yield). MS (ESI): mass calculated for $C_{26}H_{26}N_2O_3S$, 446.17; *m/z* found, 447.2 $[M + H]^+$. ^1H NMR (400 MHz, $CDCl_3$): 7.74 (d, J = 8.1, 1H), 7.66 (d, J = 7.4, 1H), 7.54 (d, J = 7.6, 2H), 7.42−7.35 (m, 3H), 7.33−7.22 (m, 4H), 6.99 (d, J = 9.0, 2H), 4.19 (t, J = 5.8, 2H), 2.98−2.86 (m, 4H), 2.65 (dt, J = 13.8, 2.1, 2H), 2.24 (dt, J = 13.4, 4.5, 2H), 1.80 (dd, J = 14.1, 2.2, 2H). Anal. ($C_{26}H_{26}N_2O_3S \cdot 0.6H_2O$) C, H, N.

1-{2-[4-(Benzothiazol-2-yloxy)phenoxy]ethyl}piperidine-4-carboxylic Acid Amide (11h). A suspension of **15** (200 mg, 0.6 mmol), isonipecotamide (73 mg, 0.6 mmol), and Silicycle dimethylamine resin (800 mg, 1.1 mmol) in CH_3CN was heated to 80 °C for 18 h. The reaction mixture was filtered, and the collected resin was rinsed with CH_3CN. The combined filtrates were concentrated under reduced pressure yielding a crude solid, which was purified on SiO_2 (10 g, 0−100% 10% [2 M NH_3 in CH_3OH] in CH_2Cl_2/CH_2Cl_2) to provide 142 mg (0.4 mmol, 63% yield) of a white solid. HRMS (ESI): mass calculated for $C_{21}H_{23}N_3O_3S$, 398.1533; *m/z* found, 398.1542 $[M + H]^+$. ^1H NMR (400 MHz, $CDCl_3$): 7.85 (dd, J = 8.0, 0.3, 1H), 7.75 (dd, J = 8.0, 0.6, 1H), 7.42 (dd, J = 7.4, 1.1, 1H), 7.32−7.22 (m, 3H), 7.02−6.91 (m, 2H), 5.67 (br d, J = 4.7, 2H), 4.15 (t, J = 5.8, 2H), 3.09 (br d, J = 8.8, 2H), 2.85 (t, J = 5.7, 2H), 2.28−2.12 (m, 3H), 2.00−1.88 (m, 2H), 1.87−1.72 (m, 2H).

(1-{2-[4-(Benzothiazol-2-yloxy)phenoxy]ethyl}piperidin-4-yl)-carbamic Acid *tert*-Butyl Ester (11i). To a mixture of **15** (98 mg, 0.3 mmol) and K_2CO_3 (92 mg, 0.7 mmol) in CH_3CN (2 mL) was added piperidin-4-ylcarbamic acid *tert*-butyl ester (74 mg, 0.4 mmol). The mixture was stirred at 70 °C for 20 h. The mixture was filtered and concentrated under reduced pressure to give a clear golden oil. The crude oil was purified on SiO_2 (4 g, 50% acetone/CH_2Cl_2) to give 101 mg of a white solid (0.2 mmol, 77% yield). MS (ESI): exact mass calculated for $C_{25}H_{31}N_3O_4S_1$, 469.61; *m/z* found, 470.2 $[M + H]^+$. ^1H NMR (400 MHz, DMSO-d_6): 7.92 (d, J = 7.9, 1H), 7.68 (d, J = 8.1, 1H), 7.45−7.25 (m, 4H), 7.06 (d, J = 9.0, 2H), 6.78 (d, J = 6.9, 1H), 4.09 (s, 2H), 3.22 (br s, 1H), 2.89 (d, J = 9.2, 2H), 2.68 (br s, 2H), 2.05 (t, J = 9.9, 2H), 1.69 (d, J = 9.6, 2H), 1.38 (s, 11H).

1-(1-{2-[4-(Benzothiazol-2-yloxy)phenoxy]ethyl}piperidin-4-yl)pyrrolidin-2-one (11j). The title compound was prepared according to the procedure for **11i** using 1-piperidin-4-yl-pyrrolidin-2-one hydrochloride[71] to provide 884 mg of **11j** as a tacky off-white solid (1.9 mmol, 99% yield). MS (ESI): mass calculated for $C_{24}H_{27}N_3O_3S$, 337.18; *m/z* found, 348.5 $[M + H]^+$. ^1H NMR (400 MHz, $CDCl_3$): 7.85 (dd, J = 8.0, 0.5, 1H), 7.75 (dd, J = 8.0, 0.8, 1H), 7.41 (dt, J = 7.3, 1.5, 1H), 7.34−7.22 (m, 3H), 7.02−6.92 (m, 2H), 4.15 (br d, J = 48.8, 2H), 3.80−3.65 (m, 1H), 3.40 (t, J = 7.0, 1H), 3.30−3.10 (br s, 1H), 3.15 (q, J = 7.2, 1H), 2.96 (br s, 1H), 2.42 (t, J = 7.9, 2H), 2.10−1.99 (m, 1H), 1.81−1.70 (m, 1H), 1.68−1.52 (m, 4H), 1.50 (d, J = 6.5, 3H). Anal. ($C_{24}H_{27}N_3O_3S$) C, H, N.

1′-{2-[4-(Benzothiazol-2-yloxy)phenoxy]ethyl}[1,4′]bipiperidinyl-2-one (11k). The title compound was prepared according to the procedure for **11i** using [1,4′]bipiperidinyl-2-one hydrochloride to provide 294 mg of **11k** as a white solid (0.7 mmol, 42% yield). MS (ESI): mass calculated for $C_{25}H_{29}N_3O_3S$, 451.19; *m/z* found, 452.4 $[M + H]^+$. ^1H NMR (400 MHz, DMSO-d_6): 7.92 (d, J = 1.1, 2H), 7.90 (d, J = 1.1, 2H), 7.42 (t, J = 7.3, 2H), 7.37 (d, J = 9.0, 2H), 7.31 (t, J = 7.3 2H), 7.06 (d, J = 9.0, 2H), 4.32−4.21 (m, 1H), 4.10 (t, J = 5.7, 2H), 3.15 (t, J = 5.3, 2H), 3.00 (d, J = 11.5, 2H), 2.71 (t, J = 5.7, 2H), 2.21 (t, J = 6.5, 2H), 2.10 (t, J = 11.4, 2H), 1.75−1.58 (m, 6H), 1.43 (d, J = 10.0, 2H). Anal. ($C_{25}H_{29}N_3O_3S$) C, H, N.

2-(1-Benzylpiperidin-4-ylamino)ethanol (17). To a solution of 1-benzyl-4-piperidinone, **16** (10.3 g, 54 mmol), and ethanolamine (13.2 mL, 218 mmol) in CH_3OH (20 mL) were added sodium cyanoborohydride (10.2 g, 163 mmol) and trifluoromethanesulfonic acid (5 mL), and the mixture was stirred at 23 °C for 3 days. The mixture was cooled to 0 °C, and 12 N HCl was slowly added until gas evolution ceased. The resulting mixture was stirred for a further 3 h. The mixture was filtered and the filtrate concentrated under reduced pressure. The crude oil was redissolved in H_2O (50 mL), and the solution was made basic by the addition of 10 N NaOH. The mixture was extracted with CH_2Cl_2 (8 × 70 mL). The combined CH_2Cl_2 extracts were dried and concentrated under reduced pressure to yield 12 g of the crude product (51 mmol, 95% yield) which was used in the next step without further purification.

3-(1-Benzylpiperidin-4-yl)oxazolidin-2-one (18). A solution of **17** (3.6 g, 15 mmol) in $ClCH_2CH_2Cl$ (5 mL) was treated with carbonyl diimidazole (CDI) (2.6 g, 16 mmol) and the mixture stirred at 23 °C for 30 min. The mixture was diluted with CH_2Cl_2 (100 mL), washed with H_2O (1 × 50 mL) and saturated aqueous $NaHCO_3$ (1 × 50 mL), dried, and concentrated under reduced pressure to yield 2.85 g (11 mmol, 65%) of the crude product which was used in the next step without purification.

3-Piperidin-4-yloxazolidin-2-one Hydrochloride Salt (19). To a solution of 3-(1-benzylpiperidin-4-yl)oxazolidin-2-one (2.3 g, 9 mmol) in $ClCH_2CH_2Cl$ (40 mL) was added α-chloroethyl chloroformate (1.5 g, 11 mmol), and the mixture was heated to 100 °C for 90 min. The mixture was cooled to 23 °C and concentrated under reduced pressure. The crude residue was dissolved in CH_3OH and heated to reflux for 1 h. The mixture was cooled to 0 °C and

concentrated under reduced pressure to yield 1.9 g of 3-piperidin-4-yl-oxazolidin-2-one hydrochloride salt (11 mmol, 99% yield). MS (ESI): exact mass calculated for $C_8H_{14}N_2O_2$, 170.1; *m/z* found, 171.2 $[M + H]^+$. ^{1}H NMR (400 MHz, DMSO-d_6): 8.97 (br s, 2H), 4.27 (dd, J = 9.1, 7.8, 2H), 3.80 (tt, J = 11.8, 4.2, 1H), 3.49 (dd, J = 8.0, 6.6, 1H), 3.30 (br d, J = 12.7, 2H), 2.97 (dt, J = 12.6, 2.3, 2H), 1.90 (ddd, J = 16.6, 13.0, 4.1, 2H), 1.86−1.75 (m, 2H).

3-(1-{2-[4-(Benzothiazol-2-yloxy)phenoxy]ethyl}piperidin-4-yl)oxazolidin-2-one (11l). The title compound was prepared according to the procedure for **11e** and **9e** using 3-piperidin-4-yloxazolidin-2-one hydrochloride salt to provide 360 mg of **11l** as a white solid (0.8 mmol, 85% yield). MS (ESI): exact mass calculated for $C_{23}H_{25}N_3O_4S_1$, 439.2; *m/z* found, 440.5 $[M + H]^+$. ^{1}H NMR (400 MHz, $CDCl_3$): 7.76 (dd, J = 8.1, 0.7, 1H), 7.69 (dd, J = 8.0, 0.7, 1H), 7.42 (dt, J = 8.6, 1.3, 1H), 7.35−7.26 (m, 3H), 7.02−6.97 (m, 2H), 4.36 (dd, J = 8.7, 7.3, 2H), 4.14 (t, J = 5.7, 2H), 3.80 (ddd, J = 16.4, 11.0, 5.6, 1H), 3.60−3.53 (m, 2H), 3.12 (br d, J = 12.0, 2H), 2.87 (d, J = 5.7, 2H), 2.28 (ddd, J = 11.7, 11.6, 3.8, 2H), 1.90−1.74(m, 4H). Anal. ($C_{23}H_{25}N_3O_4S\cdot HCl\cdot 0.3H_2O$) C, H, N.

1-{2-[4-(Benzothiazol-2-yloxy)phenoxy]ethyl}piperidin-4-ylamine (11m). To a mixture of **11i** (92 mg, 0.20 mmol) in CH_2Cl_2 (2.5 mL) was added trifluoroacetic acid (2.5 mL). The mixture was stirred at room temperature for 30 min, and CH_2Cl_2 (15 mL) was added followed by saturated $NaHCO_3$ (15 mL). The organic layer was separated, dried ($MgSO_4$), and concentrated under reduced pressure to give 60 mg of a white solid (0.20 mmol, 83% yield). MS (ESI): exact mass calculated for $C_{20}H_{23}N_3O_2S_1$, 369.49; *m/z* found, 370.2 $[M + H]^+$. ^{1}H NMR (400 MHz, DMSO-d_6): 7.92 (d, J = 7.7, 1H), 7.68 (d, J = 7.9, 1H), 7.45−7.25 (m, 4H), 7.06 (d, J = 9.0, 2H), 6.05 (br s, 2H), 4.11 (t, J = 5.6, 2H), 2.92 (d, J = 11.5, 2H), 2.81 (m, 1H), 2.71 (t, J = 5.6, 2H), 2.08 (t, J = 11.2, 2H), 1.78 (d, J = 10.7, 2H), 1.40 (m, 2H). Anal. ($C_{20}H_{23}N_3O_2S\cdot CH_2Cl_2$) C, H, N.

4-(2-Bromoethyl)phenol (21). 4-(2-Hydroxyethyl)phenol, **20** (50 g, 362 mmol), was dissolved in 48 wt % HBr (250 mL). This light-yellow solution was heated to 80 °C and stirred for 16 h. The reaction mixture was allowed to cool to room temperature and was then extracted with CH_2Cl_2 (3 × 50 mL). The combined extracts were dried, filtered, and concentrated under reduced pressure to afford 72 g (358 mmol, 100% crude yield) of a tan solid. ^{1}H NMR (400 MHz, $CDCl_3$): 9.25 (s, 1H), 7.04 (d, J = 8.4, 2H), 6.67 (d, J = 8.4, 2H), 3.62 (t, J = 7.4, 2H), 2.97 (t, J = 7.4, 2H).

2-[4-(Benzothiazol-2-yloxy)phenyl]ethanol (22). A solution of 4-(2-hydroxyethyl)phenol (867 mg, 6.3 mmol) and 2-chlorobenzothiazole 0.82 mL, 6.3 mmol) in CH_3CN was treated with finely powdered Cs_2CO_3 (4.1 g, 12.5 mmol), and the resulting suspension was stirred for 40 h at 70 °C. The reaction mixture was filtered through diatomaceous earth, and the filtrate was concentrated to a crude oil, which was purified on SiO_2 (40 g, 0−50% ethyl acetate/hexanes) to provide 940 mg (3.5 mmol, 55% yield) of a clear oil. MS (ESI): mass calculated for $C_{15}H_{13}NO_2S$, 271.07; *m/z* found, 272.2 $[M + H]^+$. ^{1}H NMR (400 MHz, $CDCl_3$): 7.78 (dd, J = 7.9, 0.4, 1H), 7.75 (dd, J = 7.9, 0.7, 1H), 7.46 (dt, J = 7.4, 1.3, 1H), 7.38−7.29 (m, 3H), 3.93 (q, J = 6.4, 2H), 2.94, (t, J = 6.5, 2H), 1.50 (m, 1H).

[4-(Benzothiazol-2-yloxy)phenyl]acetic Acid Methyl Ester (23). A solution of (4-hydroxyphenyl)acetic acid methyl ester (11.2 g, 62 mmol) and 2-chlorobenzothiazole (9.5 g mL, 56 mmol) in CH_3CN was treated with finely powdered Cs_2CO_3 (27 g, 84 mmol), and the resulting mixture was stirred at 40 °C for 17 h and 60 °C for 2 h. The reaction mixture was filtered through diatomaceous earth, and the filtrate was concentrated under reduced pressure. The crude solid was purified by dissolving in ethyl acetate (350 mL) and washing with 10% NaOH (3 × 30 mL), 0.5 M citric acid (1 × 30 mL), saturated aqueous $NaHCO_3$ (1 × 30 mL), brine (1 × 30 mL), then dried over Na_2SO_4, filtered, and concentrated under reduced pressure to provide 16 g (59 mmol, 95% yield) as a white solid. MS (ESI): mass calculated for $C_{16}H_{13}NO_3S$, 299.35; *m/z* found, 300.0 $[M + H]^+$. ^{1}H NMR (400 MHz, $CDCl_3$): 7.78 (d, J = 8.1 Hz, 1H), 7.72 (d, J = 8.1 Hz, 1H), 7.46−7.35 (m, 5H), 7.34−7.29 (m, 1H), 3.76 (s, 3H), 3.71 (s, 2H).

4-(2-Piperidin-1-ylethyl)phenol (24b). To a stirring solution of **17** (4.5 g, 22.3 mmol) in CH_3CN (100 mL) was added piperidine (3.3 mL, 33.5 mmol), followed by *N*,*N*-diisopropylethylamine (5.8 mL, 33.5 mmol). The resulting solution was stirred at 60 °C for 16 h. The mixture was allowed to cool to room temperature and concentrated under reduced pressure to yield a light-orange solid. The solid was triturated with ether (200 mL), filtered, and dried to give 4.5 g of the title compound as a white solid (21.9 mmol, 98% crude yield). MS (ESI): mass calculated for $C_{13}H_{19}NO$, 205.2; *m/z* found, 206.2 $[M + H]^+$. ^{1}H NMR (500 MHz, $CDCl_3$): 7.04−7.01 (m, 2H), 6.73−6.71 (m, 2H), 2.79−2.75 (m, 2H), 2.59−2.53 (m, 7H), 1.69−1.65, (m, 4H), 1.49−1.48 (m, 2H).

2-[4-(2-Bromoethyl)phenoxy]benzothiazole (25). A solution of **22** (174 mg, 0.6 mmol) in benzene (3 mL) was treated with PBr_3 (0.060 mL, 0.6 mmol), and the resulting suspension was heated to 70 °C for 90 min. The reaction mixture was cooled and diluted with ethyl acetate (30 mL). This solution was washed with H_2O (10 mL) and then brine (10 mL), dried, and concentrated under reduced pressure. The crude product was purified on SiO_2 (12 g, 0−50% ethyl acetate/hexanes) to provide 120 mg (0.4 mmol, 67% yield) of a light-yellow oil. MS (ESI): mass calculated for $C_{15}H_{12}BrNOS$, 332.98; *m/z* found, 335.2 $[M + H]^+$. ^{1}H NMR (400 MHz, $CDCl_3$): 7.79 (dd, J = 8.0, 0.3, 1H), 7.76 (dd, J = 8.0, 0.6, 1H), 7.42 (dt, J = 7.4, 1.2, 1H), 7.38−7.29 (m, 3H), 3.62 (t, J = 7.5, 2H), 3.25 (t, J = 7.5, 2H).

[4-(Benzothiazol-2-yloxy)phenyl]acetaldehyde (26). A solution of **23** (5.4 g, 18 mmol) in 80 mL of toluene at −90 °C was treated by dropwise addition of a 1.0 M solution of diisobutylaluminum hydride in hexanes (27 mL, 27 mmol). The mixture was slowly warmed to −68 °C over 30 min and then quenched by the addition of methanol (2.0 mL). The reaction mixture was warmed to −20 °C and diluted with diethyl ether (100 mL) and 2.0 M HCl (60 mL) and stirred vigorously for 30 min. The organic layer was separated, washed with saturated aqueous $NaHCO_3$, dried over Na_2SO_4, filtered, and concentrated to yield the title compound. This compound is best stored at low temperature in benzene and is carried on to subsequent reactions without further purification.

2-[4-(2-Piperidin-1-ylethyl)phenoxy]benzothiazole (27b). To a stirring solution of **24b** (500 mg, 2.4 mmol) in DMF (8 mL) were added Cs_2CO_3 (1.65 g, 5.1 mmol) and 2-chlorobenzothiazole (0.38 mL, 2.9 mmol). The suspension was stirred at 100 °C overnight. The reaction mixture was allowed to cool to room temperature and was then filtered through diatomaceous earth. The filtrate was concentrated under reduced pressure, and the residue was purified on SiO_2 (40 g, 0−100% acetone/CH_2Cl_2) to give 590 mg (1.7 mmol, 72% yield) of a yellow oil. MS (ESI): mass calculated for $C_{20}H_{22}N_2OS$, 338.48; *m/z* found, 339.3 $[M + H]^+$. ^{1}H NMR (400 MHz, CD_3OD): 7.76−7.73 (m, 1H), 7.64−7.61 (m, 1H), 7.42−7.25 (m, 6H), 2.88−2.83 (m, 2H), 2.60−2.52 (m, 6H), 1.66−1.61 (m, 4H), 1.53−1.45 (m, 2H). Anal. ($C_{20}H_{22}N_2OS$) C, H, N.

1-{2-[4-(Benzothiazol-2-yloxy)phenyl]ethyl}piperidin-4-ol (27e). The title compound was prepared according to the procedure for **24b** and **27b** using 4-hydroxypiperidine to provide 624 mg of **27e** as a pale oil (1.8 mmol, 88% yield). MS (ESI): mass calculated for $C_{20}H_{22}N_2O_2S$, 354.14; *m/z* found, 355.3 $[M + H]^+$. ^{1}H NMR (400 MHz, $CDCl_3$): 7.73 (d, J = 8.2, 1H), 7.64 (d, J = 8.0, 1H), 7.37 (t, J = 8.2, 1H), 7.29−7.20 (m, 5H), 3.75−3.65 (m, 1H), 2.92−2.79 (m, 4H), 2.76 (br s, 1H), 2.65−2.55 (m, 2H), 2.21 (t, J = 10.0, 2H), 1.98−1.87 (m, 2H), 1.70−1.57 (m, 2H). Anal. ($C_{20}H_{22}N_2O_2S$) C, H, N.

{1-[4-(Benzothiazol-2-yloxy)benzyl]piperidin-4-yl}carbamic Acid *tert*-Butyl Ester (27i). A solution of **26** (2.7 g, 10 mmol) and 4-BOC-aminopiperidine (2.2 g, 11 mmol) in DCE (40 mL) containing CH_3OH (0.5 mL) was treated with sodium triacetoxyborohydride (2.8 g, 13 mmol), and the mixture was stirred at room temperature for 16 h. The mixture diluted with CH_2Cl_2 (50 mL) and washed with saturated aqueous $NaHCO_3$ (15 mL), dried ($MgSO_4$), and concentrated under reduced pressure to give a white

solid. The solid was washed with diethyl ether and air-dried to give 214 mg of the product as a white solid (2.3 mmol, 23% yield). MS (electrospray): exact mass calculated for $C_{25}H_{31}N_3O_3S_1$, 453.2; *m/z* found, 454.4 $[M + H]^+$. ^1H NMR (400 MHz, $CDCl_3$): 7.73 (dd, *J* = 8.2, 0.6 Hz, 1H), 7.65 (dd, *J* = 7.9, 0.7 Hz, 1H), 7.39 (dt, *J* = 7.4, 1.3 Hz, 1H), 7.29−7.23 (m, 5H), 4.47 (br d, *J* = 6.6 Hz, 1H), 3.55−3.45 (m, 1H), 2.93 (br d, *J* = 11.3 Hz, 2H), 2.82 (dd, *J* = 11.0, 7.7 Hz, 2H), 2.60 (dd, *J* = 8.6, 5.3 Hz, 2H), 2.16 (t, *J* = 11.3 Hz, 2H), 1.97 (br. d, *J* = 11.1 Hz, 2H), 1.45 (s, 9H).

1-{2-[4-(Benzothiazol-2-yloxy)phenyl]ethyl}piperidine-4-carboxylic Acid Amide (27h). The title compound was prepared according to the procedure for **11h** using **25** and isonipecotamide to provide 56 mg of **27h** as an off-white solid (0.2 mmol, 20% yield). MS (ESI): mass calculated for $C_{21}H_{23}N_3O_2S$, 381.15; *m/z* found, 382.3 $[M + H]^+$. ^1H NMR (400 MHz, $CDCl_3$): 7.78 (dd, *J* = 8.1, 0.4, 1H), 7.71 (dd, *J* = 7.9, 0.7, 1H), 7.43 (dt, *J* = 7.5, 2.3, 1H), 7.35−7.25 (m, 5H), 5.51 (br d, *J* = 26.0, 1H), 3.09 (br d, *J* = 11.7, 2H), 2.87 (dd, *J* = 8.3, 7.6, 2H), 2.65 (dd, *J* = 8.5, 5.4, 2H), 2.29−2.18 (m, 1H), 2.13 (t, *J* = 11.4, 2H), 1.98 (br d, *J* = 11.2, 2H), 1.87−1.77 (m, 2H). Anal. ($C_{21}H_{23}N_3O_2S \cdot 0.25H_2O$) C, H, N.

1-(1-{2-[4-(Benzothiazol-2-yloxy)phenyl]ethyl}piperidin-4-yl)pyrrolidin-2-one (27j). The title compound was prepared according to the procedure for **11h** using **25** and 1-piperidin-4-ylpyrrolidin-2-one to provide 98 mg of **27j** as a white solid (0.2 mmol, 31% yield). MS (ESI): mass calculated for $C_{24}H_{27}N_3O_2S$, 421.18; *m/z* found, 422.4 $[M + H]^+$. ^1H NMR (400 MHz, $CDCl_3$): 7.76 (d, *J* = 8.0, 1H), 7.69 (dd, *J* = 8.0, 0.9, 1H), 7.41 (dt, *J* = 7.5, 1.2, 1H), 7.32−7.27 (m, 5H), 4.05 (dt, *J* = 11.9, 4.5, 1H), 3.40 (t, *J* = 7.0, 2H), 3.10 (br d, *J* = 11.7, 2H), 2.86 (dd, *J* = 11.0, 7.7, 2H), 2.66 (dd, *J* = 8.8, 5.3, 2H), 2.43 (t, *J* = 8.1, 2H), 2.19 (dt, *J* = 11.6, 2.8, 2H), 2.09−2.00 (m, 2H), 1.85−1.69 (m, 4H). The title compound was dissolved in CH_2Cl_2 and treated with 1 M HCl (1.5 equiv) in Et_2O to give the HCl salt. Anal. ($C_{24}H_{27}N_3O_2S \cdot HCl$) C, H, N.

3-(1-{2-[4-(Benzothiazol-2-yloxy)phenyl]ethyl}piperidin-4-yl)oxazolidin-2-one (27l). The title compound was prepared according to the procedure for **27i** using **19** and **26** to provide 249 mg of **27l** as an off-white solid (0.5 mmol, 92% yield). MS (ESI): exact mass calculated for $C_{23}H_{25}N_3O_3S_1$, 423.2; *m/z* found, 424.4 $[M + H]^+$. ^1H NMR (400 MHz, $CDCl_3$): 7.77 (dd, *J* = 8.1, 0.5, 1H), 7.69 (dd, *J* = 7.9, 1.1, 1H), 7.41 (dt, *J* = 8.5, 1.2, 1H), 7.35−7.28 (m, 5H), 4.36, (t, *J* = 7.8, 2H), 3.72 (t, *J* = 4.8, 2H), 3.86−3.76 (m, 1H), 3.57 (t, *J* = 8.1, 2H), 3.11 (br d, *J* = 11.8, 2H), 2.86 (dd, *J* = 11.0, 7.6, 2H), 2.66 (dd, *J* = 8.6, 5.2, 2H), 2.18 (dt, *J* = 11.7, 2.7, 2H), 1.89−1.72 (m, 4H). Anal. ($C_{23}H_{25}N_3O_3S \cdot HCl \cdot 0.5H_2O$) C, H, N

1-{2-[4-(Benzothiazol-2-yloxy)phenyl]ethyl}piperidin-4-ylamine (27m). To a stirred solution of **27i** (4.0, 8.8 mmol) in 88% formic acid (40 mL) was added 5.0 N HCl (1.8 mL, 8.8 mmol). The mixture was stirred at room temperature for 7 h and concentrated under reduced pressure to give a glass, which was triturated with Et_2O and filtered to give a solid (3.6 g, 8.8 mmol, >99% crude yield). MS (electrospray): exact mass calculated for $C_{20}H_{23}N_3O_1S_1$, 353.5; *m/z* found, 354.4 $[M + H]^+$. ^1H NMR (400 MHz, DMSO-d_6): 8.22 (s, 1H), 7.90 (d, *J* = 7.9, 1H), 7.66 (d, *J* = 7.6, 1H), 7.47−7.27 (m, 5H), 3.17−3.03 (m, 3H), 2.90−2.81 (m, 2H), 2.78−2.69 (m, 2H), 2.32 (t, *J* = 11.8, 2H), 1.96 (d, *J* = 10.7, 2H), 1.71−1.59 (m, 2H). The crude solid (1.3 g) was suspended in EtOAc (100 mL) and washed with saturated aqueous $NaHCO_3$ (30 mL). The organic layer was dried (Na_2SO_4) and concentrated to yield the desired product (1.0 g, 23 mmol, 77% yield), which was used without further purification.

***N*-(1-{2-[4-(Benzothiazol-2-yloxy)phenyl]ethyl}piperidin-4-yl)-*N*-acetamide (27n).** To a solution of **27m** (213 mg, 0.6 mmol) in CH_2Cl_2 (7 mL) at room temperature was added triethylamine (0.12 mL, 0.9 mmol), followed by acetyl chloride (0.06 mL, 0.8 mmol). The resulting mixture was stirred at room temperature 1 h. The reaction mixture was dissolved in CH_2Cl_2 (20 mL), washed with saturated aqueous $NaHCO_3$ (1 × 10 mL), dried (Na_2SO_4), and concentrated under reduced pressure to yield 211 mg (0.5 mmol, 89% yield). MS (ESI): exact mass calculated for $C_{25}H_{29}N_3O_4S$, 395.2; *m/z* found, 396.3 $[M + H]^+$. ^1H NMR (400 MHz, $CDCl_3$): 7.73 (d, *J* = 8.0, 1H), 7.66 (dd, *J* = 7.9, 0.6, 1H), 7.42−7.36 (m, 1H), 7.29−7.24 (m, 5H), 5.48 (d, *J* = 7.8, 1H), 3.87−3.77 (m 1H), 2.95 (dd, *J* = 11.6, 2H), 2.83 (dd, *J* = 10.8, 7.6, 2H), 2.62 (dd, *J* = 8.5, 5.3, 2H), 2.18 (d, *J* = 11.4, 2H), 2.00−1.94 (m, 2H), 1.98 (m, 2H), 1.49 (dddd, *J* = 12.3, 12.2, 12.2, 3.6, 2H). Anal. ($C_{22}H_{25}N_3O_2S$) C, H, N.

Acetic Acid (1-{2-[4-(Benzothiazol-2-yloxy)phenyl]ethyl}piperidin-4-ylcarbamoyl)methyl Ester (27o). The title compound was prepared from **27m** according to the procedure for **33o** to provide 253 mg of **27o** of a solid (0.6 mmol, 95% yield). MS (ESI): exact mass calculated for $C_{24}H_{27}N_3O_4S$, 453.2; *m/z* found, 454.3 $[M + H]^+$. ^1H NMR (400 MHz, $CDCl_3$): 7.73 (dd, *J* = 8.2, 0.6, 1H), 7.67 (dd, *J* = 7.9, 0.7, 1H), 7.42−7.36 (m, 1H), 7.29−7.24 (m, 5H), 6.05 (d, *J* = 8.0, 1H), 4.54 (s, 2H), 3.96−3.85 (m 1H), 3.00 (d, *J* = 8.2, 0.6, 2H), 2.85 (dd, *J* = 10.6, 7.4, 2H), 2.85 (dd, *J* = 10.6, 7.4, 2H), 2.66 (dd, *J* = 8.6, 5.3, 2H), 2.23 (t, *J* = 11.4, 2H), 2.18 (s, 3H), 1.98 (d, *J* = 9.7, 2H), 1.57 (dddd, *J* = 12.4, 12.4, 12.3, 3.6, 2H). Anal. ($C_{24}H_{27}N_3O_4S$) C, H, N.

4-(Benzothiazol-2-yloxy)benzaldehyde (30). To a mixture of 4-hydroxybenzaldehyde, **28** (1.0 g, 8.2 mmol), and 2-chlorobenzothiazole (2.0 mL, 16.4 mmol) in CH_3CN (100 mL) was added Cs_2CO_3 (5.5 g, 17.2 mmol). The reaction mixture was stirred at 60 °C for 24 h. The resulting mixture was cooled to room temperature, filtered through diatomaceous earth, and concentrated under reduced pressure to yield the crude product as an orange oil. The oil was triturated with hexanes/CH_2Cl_2 (100 mL) and the solvent layer decanted and concentrated under reduced pressure to yield an orange oil which was further purified on SiO_2 (120 g, 0−50% ethyl acetate/hexanes) to give 853 mg of a white solid (3.4 mmol, 41% yield). ^1H NMR (400 MHz, $CDCl_3$): 10.1 (s, 1H), 7.97−7.78 (m, 2H), 7.78−7.70 (m, 2H), 7.60−7.50 (m, 2H), 7.48−7.38 (m, 1H), 7.36−7.30 (m, 1H).

[4-(Benzothiazol-2-yloxy)phenyl]methanol (31). To a mixture of 4-hydroxybenzyl alcohol, **29** (12 g, 97 mmol) in CH_3CN (200 mL) containing K_2CO_3 (22 g, 159 mmol) was added 2-chlorobenzothiazole (22 g, 130 mmol), and the mixture was heated to reflux for 72 h. The mixture was cooled to room temperature, filtered, and concentrated under reduced pressure to give the crude product as a golden oil. The crude oil was purified on SiO_2 (300 g, 5% acetone/CH_2Cl_2) to give 15 g of a clear, colorless oil (58 mmol, 60% yield). MS (ESI): exact mass calculated for $C_{14}H_{11}NO_2S$, 257.1; *m/z* found, 258.3 $[M + H]^+$. ^1H NMR (400 MHz, DMSO-d_6): 7.92 (d, *J* = 7.4, 1H), 7.69 (d, *J* = 8.0, 1H), 7.50−7.31 (m, 5H), 7.32 (t, *J* = 7.5, 1H), 5.32 (t, *J* = 5.7, 1H), 4.55 (d, *J* = 5.7, 2H).

2-(4-Chloromethylphenoxy)benzothiazole (32). To a mixture of **31** (11 g, 43 mmol) in CH_2Cl_2 (100 mL) containing triethylamine (9 mL, 65 mmol) at 5 °C was added dropwise over 15 min thionyl chloride (4 mL g, 55 mmol). The ice bath was removed and the mixture warmed to room temperature and stirred for 24 h. The mixture was washed once with saturated K_2CO_3 (100 mL), dried ($MgSO_4$), and concentrated under reduced pressure to give a black oil. The crude oil was purified on SiO_2 (300 g, 100% CH_2Cl_2) to give 10 g of a clear orange oil (36 mmol, 84% yield). MS (ESI): exact mass calculated for $C_{14}H_{10}ClNOS$, 275.0; *m/z* found, 276.2 $[M + H]^+$. ^1H NMR (400 MHz, DMSO-d_6): 7.95 (d, *J* = 7.3, 1H), 7.70 (d, *J* = 7.6, 1H), 7.59 (d, *J* = 8.6, 2H), 7.47 (d, *J* = 8.6, 2H), 7.37 (t, *J* = 7.4, 1H), 7.33 (t, *J* = 7.5, 1H), 4.84 (s, 2H).

2-(4-Piperidin-1-ylmethylphenoxy)benzothiazole (33b). The title compound was prepared according to the procedure for **33j**, using piperidine to provide 125 mg of **33b** as a pale-yellow solid (0.4 mmol, 47% yield). MS (ESI): mass calculated for $C_{19}H_{20}N_2OS$, 324.4; *m/z* found, 325.3 $[M + H]^+$. ^1H NMR (400 MHz, $CDCl_3$): 7.75 (d, *J* = 8.2, 1H), 7.73 (d, *J* = 8.2, 1H), 7.36−7.42 (m, 3H), 7.24−7.32 (m, 3H), 3.49 (s, 2H), 2.39 (br s, 3H), 1.56−1.64 (m, 5H), 1.40−1.48 (m, 2H). Anal. ($C_{19}H_{20}N_2OS$) C, H, N.

1-[4-(Benzothiazol-2-yloxy)benzyl]piperidin-4-ol (33e). The title compound was prepared according to the procedure for **33j** using 4-hydroxypiperidine to provide 383 mg of **33e** as a white

solid (1.1 mmol, 58% yield). MS (ESI): mass calculated for $C_{19}H_{20}N_2O_2S$, 340.4; *m/z* found, 341.3 $[M + H]^+$. ^{1}H NMR (400 MHz, $CDCl_3$): 7.74 (d, J = 8.0, 1H), 7.67 (d, J = 8.0, 1H), 7.42−7.36 (m, 3H), 7.32−7.24 (m, 3H), 3.68−3.64 (m, 1H), 3.46 (s, 2H), 2.81−2.25 (m, 2H), 2.20−2.15 (m, 2H), 1.95−1.90 (m, 2H), 1.67−1.58 (m, 3H). Anal. ($C_{19}H_{20}N_2O_2S\cdot0.3H_2O$) C, H, N.

1-[4-(Benzothiazol-2-yloxy)benzyl]piperidine-4-carboxylic Acid Amide (33h). To a mixture of **32** (150 mg, 0.5 mmol) in CH_3CN (5 mL) containing Cs_2CO_3 (373 mg, 1.1 mmol) was added isonipecotamide (139 mg, 1.1 mmol), and the mixture was heated to 70 °C for 24 h. The mixture was cooled to room temperature, filtered, and concentrated under reduced pressure to give the crude product as a pale-yellow oil. The crude oil was purified on SiO_2 (12 g, 0−10% 2 M NH_3 in CH_3OH/CH_2Cl_2) to give a white solid (82 mg, 0.2 mmol, 41% yield). MS (ESI): mass calculated for $C_{20}H_{21}N_3O_2S$, 367.1; *m/z* found, 368.3 $[M + H]^+$. ^{1}H NMR (400 MHz, DMSO-d_6): 7.94 (d, J = 7.4, 1H), 7.70 (d, J = 7.9, 1H), 7.48−7.38 (m, 5H), 7.33 (d, J = 7.1, 1H), 7.22 (s, 1H), 6.73 (s, 1H), 3.49 (s, 2H), 2.83 (d, J = 11.4, 2H), 2.14−2.02 (m, 1H), 2.00−1.93 (m, 2H), 1.73−1.62 (m, 2H), 1.58 (t, J = 15.4, 2H). Anal. ($C_{20}H_{21}N_3O_2S\cdot0.25H_2O$) C, H, N.

{1-[4-(Benzothiazol-2-yloxy)benzyl]piperidin-4-yl}carbamic Acid *tert*-Butyl Ester (33i). A mixture of **30** (500 mg, 1.9 mmol), piperidin-4-ylcarbamic acid *tert*-butyl ester (785 mg, 3.9 mmol), and molecular sieves (500 mg, crushed, 4 Å) in $ClCH_2CH_2Cl$ (10 mL) was stirred at room temperature for 40 min. To the resulting reaction mixture was added $NaBH(OAc)_3$ portionwise over 1.5 h (4 × 207 mg, 3.9 mmol). The resulting mixture was stirred at room temperature for 24 h, filtered through diatomaceous earth, and rinsed with CH_2Cl_2 (50 mL). The filtrate was washed with saturated aqueous $NaHCO_3$ (1 × 25 mL), dried (Na_2SO_4), and concentrated under reduced pressure to yield the crude product as a light-yellow oil. The crude product was purified on SiO_2 (40 g, 0−5% 2 M NH_3 in CH_3OH/CH_2Cl_2) to give a white foam (504 mg, 1.1 mmol, 59% yield). MS (ESI): mass calculated for $C_{24}H_{29}N_3O_3S$, 439.6; *m/z* found, 440.4 $[M + H]^+$. ^{1}H NMR (400 MHz, $CDCl_3$): 7.74 (d, J = 8.0, 1H), 7.67 (d, J = 8.0, 1H), 7.42−7.36 (m, 3H), 7.32−7.24 (m, 3H), 4.44 (br s, 1H), 3.50 (s, 2H), 2.82−2.76 (m, 2H), 2.16−2.06 (m, 2H), 1.96−1.88 (m, 2H), 1.45 (s, 9H), 1.48−1.38 (m, 2H).

1-{1-[4-(Benzothiazol-2-yloxy)benzyl]piperidin-4-yl}pyrrolidin-2-one (33j). A mixture of **30** (500 mg, 1.9 mmol), 1-piperidin-4-ylpyrrolidin-2-one hydrochloride (440 mg, 2.2 mmol), Et_3N (300 μL, 2.2 mmol), and molecular sieves (500 mg, crushed, 4 Å) in $ClCH_2CH_2Cl$ (10 mL) was stirred at room temperature for 1 h. To the resulting mixture was added $NaBH(OAc)_3$ (830 mg, 3.9 mmol). The mixture was stirred at room temperature for 24 h, filtered through diatomaceous earth, rinsed with CH_2Cl_2 (50 mL), and concentrated under reduced pressure to yield the crude product as a yellow oil. The crude product was purified on SiO_2 (40 g, 0−100% acetone/CH_2Cl_2) to give a clear oil which crystallized on standing (290 mg, 0.70 mmol, 36% yield). MS (ESI): mass calculated for $C_{23}H_{25}N_3O_2S$, 407.5; *m/z* found, 408.3 $[M + H]^+$. ^{1}H NMR (400 MHz, $CDCl_3$): 7.76 (d, J = 8.0, 1H), 7.69 (d, J = 8.0, 1H), 7.41−7.30 (m, 3H), 7.35−7.27 (m, 3H), 4.46−3.98 (m, 1H), 3.71 (s, 2H), 3.36 (t, J = 6.9, 2H), 2.97 (d, J = 11.7, 2H), 2.40 (t, J = 8.1, 2H), 2.17−1.97 (m, 4H), 1.81−1.62 (m, 4H). Anal. ($C_{23}H_{25}N_3O_2S\cdot0.15H_2O$) C, H, N.

3-{1-[4-(Benzothiazol-2-yloxy)benzyl]piperidin-4-yl}oxazolidin-2-one (33l). The title compound was prepared according to the procedure for **33h** using **19**and K_2CO_3 to provide 591 mg of **33l** as a golden oil (1.4 mmol, 84% yield). MS (ESI): mass calculated for $C_{22}H_{23}N_3O_3S$, 409.2; *m/z* found, 410.3 $[M + H]^+$. ^{1}H NMR (400 MHz, DMSO-d_6): 7.94 (d, J = 8.7, 1H), 7.70 (d, J = 7.5, 1H), 7.48−7.37 (m, 5H), 7.33 (d, J = 8.0, 1H), 4.25 (t, J = 7.7, 2H), 3.60−3.42 (m, 5H), 2.85 (d, J = 11.5, 2H), 2.04 (t, J = 11.4, 2H), 1.78−1.58 (m, 4H). Anal. ($C_{22}H_{23}N_3O_3S$) C, H, N.

1-[4-(Benzothiazol-2-yloxy)benzyl]piperidin-4-ylamine Dihydrochloride (33m). To a solution of **33i** (200 mg, 0.5 mmol) in CH_2Cl_2 (2 mL) at 0 °C was added 4 N HCl in dioxane (1.8 mL, 7.2 mmol) dropwise. The resulting reaction mixture was stirred at room temperature for 2 h. The desired product was isolated by filtration and was rinsed with Et_2O (50 mL) to yield a white powder (187 mg, 0.5 mmol, 100% yield). MS (ESI): mass calculated for $C_{19}H_{21}N_3OS$, 339.5; *m/z* found, 340.4 $[M + H]^+$. ^{1}H NMR (400 MHz, $CDCl_3$): 7.68−7.64 (m, 1H), 7.58−7.52 (m, 2H), 7.48−7.44 (m, 1H), 7.40−7.35 (m, 2H), 7.30−7.24 (m, 1H), 7.20−7.14 (m, 1H), 4.25 (s, 2H), 3.52−3.46 (m, 2H), 3.36−3.28 (m, 1H), 3.08−2.99 (m, 2H), 2.16−2.08 (m, 2H), 1.92−1.80 (m, 2H).

***N*-{1-[4-(Benzothiazol-2-yloxy)benzyl]piperidin-4-yl}acetamide (33n).** To a solution of **33m** (413 mg, 1.0 mmol) in CH_2Cl_2 (20 mL) at room temperature was added triethylamine (0.70 mL, 5.0 mmol), followed by acetyl chloride (0.09 mL, 1.2 mmol). The resulting mixture was stirred at room temperature overnight. The reaction mixture was dissolved in CH_2Cl_2 (100 mL), washed with saturated aqueous $NaHCO_3$ (1 × 25 mL), dried (Na_2SO_4), and concentrated under reduced pressure to yield the crude product as an off-white solid. The crude product was purified on SiO_2 (40 g, 0−10% CH_3OH/CH_2Cl_2) to give a white solid (271 mg, 0.7 mmol, 71% yield). MS (ESI): mass calculated for $C_{21}H_{23}N_3O_2S$, 381.15; *m/z* found, 382.4 $[M + H]^+$. ^{1}H NMR (400 MHz, $CDCl_3$): 7.73 (d, J = 7.8, 1H), 7.67 (d, J = 8.1, 1H), 7.43−7.36 (m, 3H), 7.33−7.24 (m, 3H), 5.65 (br s, 1H), 3.87−3.76 (m, 1H), 3.54 (s, 2H), 2.86 (d, J = 12.1, 2H), 2.17 (t, J = 11.4, 2H), 1.97 (s, 3H), 1.93 (d, J = 11.9, 2H), 1.56−1.44 (m, 2H). Anal. ($C_{21}H_{23}N_3O_2S\cdot0.25H_2O$)

Acetic Acid {1-[4-(Benzothiazol-2-yloxy)benzyl]piperidin-4-ylcarbamoyl}methyl Ester (33o). To a solution of **33m** (413 mg, 1.0 mmol) in CH_2Cl_2 (20 mL) at room temperature was added Et_3N (0.70 mL, 5.0 mmol), followed by acetoxyacetyl chloride (0.16 mL, 1.5 mmol). The resulting mixture was stirred at room temperature overnight. The reaction mixture was dissolved in CH_2Cl_2 (100 mL), washed with saturated aqueous $NaHCO_3$ (1 × 25 mL), dried (Na_2SO_4), and concentrated under reduced pressure to yield the crude product as an off-white solid. The crude product was purified on SiO_2 (40 g, 0−10% CH_3OH/CH_2Cl_2) to give a white solid (410 mg, 0.9 mmol, 93% yield). MS (ESI): mass calculated for $C_{23}H_{25}N_3O_4S$, 439.2; *m/z* found, 440.4 $[M + H]^+$. ^{1}H NMR (400 MHz, $CDCl_3$): 7.72 (d, J = 7.8, 1H), 7.65 (d, J = 8.1, 1H), 7.40−7.35 (m, 3H), 7.32−7.23 (m, 3H), 6.11 (d, J = 8.3, 1H), 4.53, (s, 2H), 3.93−3.82 (m, 1H), 3.50 (s, 2H), 2.83 (d, J = 11.9, 2H), 2.15 (s, 3H), 2.14 (t, J = 11.9, 2H), 1.93 (d, J = 12.1, 2H), 1.56−1.45 (m, 2H). Anal. ($C_{23}H_{25}N_3O_3S$) C, H, N.

***N*-{1-[4-(Benzothiazol-2-yloxy)benzyl]piperidin-4-yl}-2-hydroxyacetamide (33p).** To a solution of **33o** (370 mg, 0.84 mmol) in THF (30 mL), CH_3OH (10 mL), and H_2O (10 mL) was added lithium hydroxide (80 mg, 3.3 mmol). The resulting mixture was stirred at room temperature overnight. The mixture was extracted with CH_2Cl_2 (30 mL × 3). The combined organic phases were concentrated under reduced pressure to yield the crude product as an off-white solid. The crude product was purified on SiO_2 (40 g, 0−10% CH_3OH/CH_2Cl_2) to give a white solid (300 mg, 82% yield). MS (ESI): mass calculated for $C_{21}H_{23}N_3O_3S$, 397.2; *m/z* found, 398.3 $[M + H]^+$. ^{1}H NMR (400 MHz, $CDCl_3$): 7.72 (d, J = 7.8, 1H), 7.65 (d, J = 8.1, 1H), 7.40−7.35 (m, 3H), 7.31−7.23 (m, 3H), 6.83 (d, J = 8.1, 1H), 5.33 (br s, 1H), 3.99 (s, 2H), 3.87−3.75 (m, 1H), 3.50 (s, 2H), 2.85 (d, J = 11.4, 2H), 2.14 (t, J = 10.9, 2H), 1.92 (d, J = 12.6, 2H), 1.56−1.43 (m, 2H). Anal. ($C_{21}H_{24}N_3O_3S$) C, H, N.

1-(4-Benzyloxybenzyl)piperidine-4-carboxylic Acid Ethyl Ester (35). A mixture of **34** (15.2 g, 65.3 mmol), isonipecotic acid ethyl ester (15 mL, 97 mmol), and K_2CO_3 (13.5 g, 97.6 mmol) in CH_3CN (300 mL) was stirred at reflux for 20 h. The reaction mixture was cooled to room temperature and filtered. The solvent was removed under reduced pressure to yield a clear golden oil. This material was diluted with ^{i}PrOH (100 mL), and the mixture was filtered. The solid was air-dried to yield a white solid (19.7 g, 55.5 mmol, 85% yield). TLC (SiO_2, 15% acetone/CH_2Cl_2): R_f = 0.32. MS (ESI): mass calculated for $C_{22}H_{27}NO_3$, 353.2; *m/z* found, 354.3 $[M + H]^+$. ^{1}H NMR (400 MHz, DMSO-d_6): 7.44 (d, J = 7.1, 2H), 7.39 (t, J = 7.1, 2H), 7.33 (d, J = 7.2, 1H), 7.18 (d, J = 8.2, 2H), 6.94 (2H, J = 8.6, 2H), 5.08 (s, 2H), 4.04 (q, J = 7.09,

2H), 2.72 (d, $J = 11.5$, 2H), 2.32−2.18 (m, 1H), 1.94 (t, $J = 11.6$, 2H), 1.76 (d, $J = 10.2$, 2H), 1.59−1.48 (m, 2H), 1.17 (t, $J = 7.1$, 3H).

1-(4-Hydroxybenzyl)piperidine-4-carboxylic Acid Ethyl Ester (36). Compound **35** (10.0 g, 28.3 mmol) was dissolved in 1:1 ethanol/ethyl acetate (150 mL). To this solution was added Pd on carbon (10 wt %, 503 mg) as a suspension in ethanol (5.0 mL). The resulting suspension was placed on a Parr hydrogenator at 40 psi of H_2 and shaken overnight. The reaction mixture was filtered through a pad of diatomaceous earth, and the filtrate was concentrated under reduced pressure to give a clear golden oil. The oil was purified on SiO_2 (90 g, 50% acetone/CH_2Cl_2) to give a white solid (2.0 g, 7.6 mmol, 27% yield). TLC (SiO_2, 50% acetone/CH_2Cl_2): $R_f = 0.32$. MS (ESI): mass calculated for $C_{15}H_{21}NO_3$, 263.2; *m/z* found, 264.2 $[M + H]^+$. ^{1}H NMR (400 MHz, DMSO-d_6): 9.25 (s, 1H), 7.05 (d, $J = 8.4$, 2H), 6.68 (d, $J = 8.4$, 2H), 4.04 (q, $J = 7.1$, 2H), 3.34 (s, 2H), 2.71 (d, $J = 11.5$, 2H), 2.32−2.18 (m, 1H), 1.92 (t, $J = 11.6$, 2H), 1.76 (d, $J = 10.2$, 2H), 1.59−1.48 (m, 2H), 1.17 (t, $J = 7.1$, 3H).

1-[4-(Benzothiazol-2-yloxy)benzyl]piperidine-4-carboxylic Acid Ethyl Ester (33q). To a stirring solution of **36** (510 mg, 1.9 mmol) in CH_3CN (15 mL) were added K_2CO_3 (560 mg, 4.1 mmol) and 2-chlorobenzothiazole (0.50 mL, 4.0 mmol). The suspension was heated to 80 °C and stirred overnight. The reaction mixture was allowed to cool to room temperature and then filtered through diatomaceous earth. The filtrate was concentrated under reduced pressure, and the residue was purified on SiO_2 (12 g, 0−15% acetone/CH_2Cl_2) to give a clear and colorless tacky oil (720 mg, 1.7 mmol, 94% yield). TLC (SiO_2, 15% acetone/CH_2Cl_2): $R_f = 0.5$. MS (ESI): mass calculated for $C_{22}H_{24}N_2O_3S$, 396.2; *m/z* found, 397.3 $[M + H]^+$. ^{1}H NMR (400 MHz, DMSO-d_6): 7.92 (d, $J = 8.0$, 1H), 7.68 (d, $J = 8.0$, 1H), 7.48−7.33 (m, 5H), 7.33 (t, $J = 7.1$, 1H), 4.06 (q, $J = 7.1$, 2H), 3.49 (s, 2H), 2.76 (d, $J = 11.5$, 2H), 2.34−2.22 (m, 1H), 2.02 (t, $J = 11.6$, 2H), 1.80 (d, $J = 10.2$, 2H), 1.64−1.54 (m, 2H), 1.18 (t, $J = 7.1$, 3H).

1-[4-(Benzothiazol-2-yloxy)benzyl]piperidine-4-carboxylic Acid (33r). To a stirring solution of **33q** (663 mg, 1.7 mmol) in 25% ^{i}PrOH/H_2O (20 mL) was added potassium hydroxide (206 mg, 3.1 mmol). The reaction mixture was stirred at room temperature for 20 h, and the solution was treated to pH 5.5 with 1 M HCl. The resulting solution was extracted with 10% ^{i}PrOH/$CHCl_3$ (3 × 50 mL). The combined extracts were dried ($MgSO_4$), filtered, and concentrated under reduced pressure to yield a white solid (561 mg, 1.6 mmol, 91% yield). MS (ESI): mass calculated for $C_{20}H_{20}N_2O_3S$, 368.1; *m/z* found, 369.3 $[M + H]^+$. ^{1}H NMR (400 MHz, DMSO-d_6): 7.92 (d, $J = 7.6$, 1H), 7.69 (d, $J = 7.6$, 1H), 7.48−7.34 (m, 5H), 7.33 (t, $J = 7.1$, 1H), 3.49 (s, 2H), 2.76 (d, $J = 11.4$, 2H), 2.22−2.11 (m, 1H), 2.02 (t, $J = 11.2$, 2H), 1.80 (d, $J = 13.2$, 2H), 1.62−1.48 (m, 2H), 1.18 (t, $J = 7.1$, 3H). Anal. ($C_{20}H_{20}N_2O_3S \cdot 0.2H_2O$) C, H, N.

Biological Methods. All methods used to evaluate the biological activity of compounds described in this publication have been reported previously.[37] All in vitro assays are the average of three or more determinations unless otherwise noted.

Preparation of Recombinant Human LTA_4 Hydrolase. LTA_4H-encoding DNA was amplified by polymerase chain reaction (PCR) and cloned into pFastBac1 (Invitrogen) for expression in *Spodoptera frugiperda* (Sf-9) cells. Recombinant LTA_4H enzyme was purified from the infected Sf-9 cells as previously described[72] and adjusted to 0.29 mg/mL protein in 50 mM Tris (pH 8.0), 150 mM NaCl, 5 mM dithiothreitol, 50% glycerol, and EDTA-free complete protease inhibitor cocktail (Roche). The specific activity of the enzyme was about 3.8 (mmol/min)/mg.

Preparation of Substrate. LTA_4 substrate was prepared from the methyl ester of LTA_4 (Cayman Chemical) by treatment under nitrogen with 67 mol equiv of NaOH at room temperature for 40 min. The LTA_4 substrate in its free acid form was kept frozen at −80 °C until needed.

Epoxide Hydrolase Assay. LTA_4 hydrolase inhibitors were prepared as 10 mM stock solutions in DMSO and diluted in the assays so that the final DMSO concentration did not exceed 0.1%. Recombinant human LTA_4H (36 ng) was incubated with various concentrations of test compound for 10 min at room temperature in assay buffer (0.1 M potassium phosphate, pH 7.4, 5 mg/mL fatty acid-free bovine serum albumin) in a volume of 50 μL. The solution was then adjusted to 200 μL with assay buffer, and 25 μL of the substrate, LTA_4, was added (final concentration of 40 ng/mL, 0.13 μM, final volume of 225 μL). After 10−30 min at room temperature, the assay was terminated by diluting 20-fold in assay buffer. The amount of LTB_4 produced was assayed by enzyme immunoassay (EIA) (Assay Designs, Inc., catalog no. 901-068). The concentration of compound that was required for half-maximal inhibition of recombinant enzyme activity (IC_{50}) was calculated by nonlinear regression using Graphpad Prism 4.0, one site binding competition.

[^{3}H]Astemizole Binding Assay. Compounds were assessed for their ability to displace [^{3}H]astemizole using a modification of methods previously described.[73–75] Membranes from HEK-293 cells expressing the hERG K^+ channel and the [*O*-methyl-^{3}H]astemizole ligand were purchased from Perkin-Elmer (Boston, MA). Assays were carried out in 96-well v-bottom plates in a total volume of 250 μL containing 17 μg of membrane and 2 nM final assay concentration (FAC) of [^{3}H]astemizole in a binding buffer composed of 10 mM HEPES, 40 mM KCl, 20 mM KH_2PO_4, 0.5 mM $KHCO_3$, 5 mM $MgCl_2$, 10 mM glucose, 50 mM glutamic acid, 0.02 mM aspartic acid, 1 mM EGTA, and 0.1% BSA. Test compounds were added in 2.5 μL of 100% DMSO (1% FAC). Nonspecific binding was determined using 10 μM terfenadine (FAC). Assays were allowed to come to equilibrium for 60 min at room temperature, and the assay was terminated by rapid filtration through 96-well GF/B filter plates (Perkin-Elmer) presoaked in 0.3% polyethylemeimine (PEI). The filter plates were washed four times with an ice cold solution of 25 mM Tris, 130 mM NaCl, 0.8 mM $MgCl_2$, 0.05 mM $CaCl_2$, 5.5 mM KCl, 5 mM glucose, and 0.1% BSA and dried overnight at room temperature, and 30 mL of Microscint 0 (Perkin-Elmer) was added prior to reading in a TopCount NXT (Perkin-Elmer).

In Vivo Experiments. All animal experiments described in this study were performed after review of the protocols and approval by the Institutional Animal Care and Use Committee.

Murine Whole Blood Leukotriene B_4 Assay. For in vitro assays of ionophore-stimulated lipid mediator production, CD-1 mice were euthanized and blood was collected in heparin-containing syringes by cardiac puncture. The blood was diluted 1:15 in RPMI-1640 medium, and 200 μL aliquots of the diluted blood were added to wells of a 96-well tissue culture plate. Test compounds were added at different concentrations to the diluted whole blood (final DMSO concentration of 0.1%) and preincubated for 15 min at 37 °C in a humidified incubator. For murine ex vivo analysis of LTB_4 production, blood was obtained from Balb/c mice 4 h after oral dosing of test compound and was diluted 1:1 in RPMI-1640 medium, after which 200 μL aliquots of the diluted blood were added to a microtiter plate. Calcium ionophore A23187 (Sigma Chemical Co., St. Louis, MO) was added to samples for both in vitro and ex vivo whole blood assays (final concentration of 20 μg/mL). The incubation was continued under the same conditions for an additional 10−30 min to allow eicosanoid formation. The reaction was terminated by centrifugation (208*g*, 10 min at 4 °C) to form a cell pellet, and the amount of LTB_4 produced was assayed in the supernatants (diluted $^1/_5$ to $^1/_{15}$) by enzyme immunoassay. According to the manufacturer, the LTB_4 EIA assay is selective for LTB_4, with 5% cross-reactivity for 6-*trans*-12-epi-LTB_4 and 6-*trans*-LTB_4. IC_{50} values were determined as described above for epoxide hydrolase activity.

Arachidonic Acid Induced Ear Inflammation. Arachidonic acid (Calbiochem catalog no. 181198) in a stock solution of 1 g/mL was diluted to 100 mg/mL in 100% acetone for experimental use.

Test compounds or vehicle, 20% hydroxypropyl-β-cyclodextrun, HPβCD, were administered orally at $t = -1$ h to female BALB/c mice. At $t = 0$, 2 mg of arachidonic acid in acetone was applied to one ear of each mouse ($n = 8-10$ per group) and acetone to the other ear. At $t = 3$ h, the mice were sacrificed, blood was drawn

for ex vivo whole blood LTB_4 production assays and measurement of compound levels. At the same time, 8 mm ear biopsies were taken, weighed, and frozen at −80 °C for future analysis of neutrophil influx, as measured by myeloperoxidase activity.

Myeloperoxidase Assay. Ear biopsies were thawed and roughly minced into FastPrep tubes with lysing matrix D (Q-Biogene catalog no. 6913-100) prior to addition of 0.5 mL of freshly made extraction buffer (0.3 M sucrose, 0.22% cetyltrimethylammoniumbromide (CTAB), 2.5 mM citrate). Samples were run on the FastPrep homogenization instrument for 30 s at a speed of 5 m/s prior to storage on ice. Samples were then centrifuged for 10 min at 14 000 rpm in a microfuge. In a 96-well plate, 10 μL of the resultant supernatant was added to 90 μL of freshly made dilution buffer (10 mM citrate, pH 5.0, 0.22% CTAB). Tetramethylbromidine (TMB) (20 μL) was added, and the plate was mixed gently and incubated at room temperature for 1 h. The reaction was then stopped with 100 μL of 1 M H_2SO_4, and the plate was read in a spectrophotometric plate reader at 405 nm.

TNBS Colitis Model. The colitis model and analysis were performed as previously described.[76] Briefly, colitis was induced in male Wistar rats (200−240 g) by intracolonic instillation of 10 mg of trinitrobenzenesulfonic acid (TNBS) administration on day 0. At the end of the experiment, 72 h after TNBS administration the distal colon was exposed and the terminal 8 cm dissected, photographed, and stored appropriately for subsequent analysis of % lesion area and biochemical parameters. In three separate experiments animals were dosed with either vehicle (20% HPβCD), **33p**, **33r**, or **10b**, at 30 mg/kg b.i.d., commencing 24 h prior to the induction of disease. There were 10 animals per group, including one group that received no TNBS, therefore acting as a nondiseased baseline control. Consequently, % inhibition of % colonic lesion area, tissue myeloperoxidase, and LTB_4 for each compound was calculated as follows: [(test article response) − (basal response)]/[(vehicle response) − (basal response)] × 100.

Pharmacokinetic Analysis in Rats and Dogs. Compound, prepared in 20% HPβCD, was administered intravenously (2 or 3 mg/kg) or orally (10 or 30 mg/kg) to groups of three male rats or female beagle dogs. At various time intervals, blood was removed into heparin-containing syringes and the plasma levels of the dosed compound were determined by LC/MS analysis relative to standard curves generated in the same matrix. The pharmacokinetic parameters C_{max} (maximum plasma concentration), t_{max} (time of maximum plasma concentration), $AUC_{0-\infty}$ (area under the plasma concentration time curve), $t_{1/2}$ (half-life in plasma), CL/*F* (total clearance over bioavailable fraction), Vd_{ss} (volume of distribution at steady state), and *F* (bioavailable fraction) were determined from the data using WinNonLin software (Pharsight, Mountain View, CA).

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Supporting Information Available: Combustion analysis data for compounds **10a**−**e**, **11a**−**g**, **6j**−**m**, **8a**−**d**, **27b**, **27e**, **27h**, **27j**, **27l**, **27n**, **27o**, **33b**, **33e**, **33h**, **33j**, **33n**, **33o**, **33p**, **33r** and selectivity data for **10b**, **33p**, and **33r**. This material is available free of charge via the Internet at http://pubs.acs.org.

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(51) Accelrys Software Inc., San Diego, CA. The crystal structure of LTA4H complexed with bestatin was used for docking.
(52) The compound 1-[2-(4-benzyloxyphenoxy)ethyl]piperidine has a reported enzymatic IC_{50} value of 30 nM and an IC_{50} value of 310 nM in human whole blood. This compound in our assays had an enzymatic IC_{50} value of 38 nM and an IC_{50} of 343 nM in mouse whole blood.
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(59) Data generated by Absorption Systems LP, Exton, PA 19341 ($t_{1/2}$: human LM > 100 min, rat S9 > 100 min, mouse S9 = 40 min, dog S9 = 54 min). These data provide an early indicator of pharmcokinetic behavior and potential for first pass metabolism across tested species. The reaction mixture, minus the test compound, was prepared using 0.5 or 1 mg/mL microsomes, 1 mM NADPH, 100 mM potassium phosphate (pH 7.4), and magnesium chloride (10 mM) and equilibrated at 37 °C for 3 min. The reaction was initiated by addition of the test compound (5 μM) and then incubated in a water bath at 37 °C. Aliquots (100 mL) were withdrawn in triplicate at 0, 30, and 60 min and combined with 400 mL of ice-cold 50/50 acetonitrile/H_2O to terminate the reaction. Several controls (testosterone, propranolol, and atenolol) were run simultaneously with the test compounds. The percent compound remaining in the incubation mixture was plotted as a function of time; a first-order exponential equation was fit to the observed data. The elimination half-lives associated with the disappearance of test and control compounds were determined to compare their relative metabolic stability.
(60) A related compound, **29r**, did not show a reduction in the formation of LTC4 when dosed orally, supporting that these compounds are not interacting with 5-LO or FLAP.
(61) Selectivity of **5b**, **29p**, and **29r** was evaluated by Cerep, Inc. (Paris, France) against 50 other targets. These targets represent major classes of biogenic amine receptors, neuropeptide receptors, ion channel binding sites, and neurotransmitter transporters. Selectivity of **5b** was evaluated by Cerep, Inc. (Paris, France) against 24 other enzyme targets. Details are included in the Supporting Information. Microsomal stability of **29p** was also evaluated by Cerep, Inc. (Paris, France).
(62) Data generated by Absorption Systems LP, Exton, PA 19341, at t = 24 h under equilibrium conditions using phosphate buffers of described pH.
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