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Characterization of Tunable Piperidine and Piperazine Carbamates as Inhibitors of Endocannabinoid Hydrolases

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Monoacylglycerol lipase (MAGL) and fatty acid amide hydrolase (FAAH) are two enzymes from the serine hydrolase superfamily that degrade the endocannabinoids 2-arachidonoylglycerol and anandamide, respectively. We have recently discovered that MAGL and FAAH are both inhibited by carbamates bearing an *N*-piperidine/piperazine group. Piperidine/piperazine carbamates show excellent in vivo activity, raising brain endocannabinoid levels and producing CB1-dependent behavioral effects in mice, suggesting that they represent a promising class of inhibitors for studying the endogenous functions of MAGL and FAAH. Herein, we disclose a full account of the syntheses, structure−activity relationships, and inhibitory activities of piperidine/piperazine carbamates against members of the serine hydrolase family. These scaffolds can be tuned for MAGL-selective or dual MAGL-FAAH inhibition by the attachment of an appropriately substituted bisarylcarbinol or aryloxybenzyl moiety, respectively, on the piperidine/piperazine ring. Modifications to the piperidine/piperazine ring ablated inhibitory activity, suggesting a strict requirement for a six-membered ring to maintain potency.

The endogenous cannabinoid system is composed of the G-protein-coupled receptors CB1 and CB2, their endogenous ligands anandamide (AEA[a]) and 2-arachidonoylglycerol (2-AG) (the "endocannabinoids"), and the enzymes that biosynthesize and degrade endocannabinoids.[1] The magnitude and duration of brain endocannabinoid signaling are tightly regulated by enzymatic hydrolysis, a process that involves distinct enzymes for each endocannabinoid. Fatty acid amide hydrolase (FAAH) is the principal hydrolytic enzyme for AEA.[2] Genetic or pharmacological disruption of FAAH in rodents causes dramatic elevations (> 10-fold) in brain AEA levels and CB1-dependent analgesic responses in several acute and chronic pain models.[3−5] Although several enzymes can hydrolyze 2-AG in vitro, this activity appears to be principally mediated by monoacylglycerol lipase (MAGL) in the rodent brain.[6,7] Recently, we described a highly selective and efficacious MAGL inhibitor **1** (JZL184, Figure 1), a piperidine carbamate that, upon administration to mice, reduces brain 2-AG hydrolysis activity, elevates total brain 2-AG levels by > 8-fold, and causes CB1-dependent hypomotility and analgesia.[7,8] We have also described **2** (JZL195, Figure 1), a dual FAAH-MAGL inhibitor based on a piperazine carbamate scaffold, that elevates total brain AEA and 2-AG to levels comparable to selective inhibitors of each individual enzyme.[9] Interestingly, **2** produces strong antinociceptive and cataleptic effects that provide evidence for crosstalk between AEA and 2-AG signaling pathways in vivo. Concurrently with these studies, highly selective inhibitors of FAAH have been introduced that use a piperazine or piperidine urea to inactivate this enzyme. Examples include **3** (PF-622) and **4** (PF-3845) (Figure 1).[5,10] Collectively, these data indicate that piperazine/piperidine carbamates and ureas may represent privileged chemical scaffolds[11,12] for generating covalent, selective, and efficacious inhibitors of endocannabinoid hydrolases. Herein, we provide a full account of the discovery, synthesis, structure−activity relationships, and inhibitory activities of compounds that led to the identification of **1** and **2** and we show that by attachment of an appropriately substituted 4-bisarylcarbinol or 4-aryloxybenzyl moiety, the versatile piperidine/piperazine carbamate scaffold can be tuned to generate MAGL-selective or dual FAAH-MAGL inhibitors.

Results and Discussion

Lead Endocannabinoid Hydrolase Inhibitors Discovered by Competitive Activity-Based Protein Profiling (ABPP). Both FAAH and MAGL are members of the serine hydrolase superfamily of enzymes that use a conserved serine nucleophile for catalysis.[13−15] Our search for selective inhibitors of MAGL or dual inhibitors for FAAH-MAGL has therefore benefited from some unusual features of this family. First, the catalytic serine is susceptible to covalent inactivation by several electrophilic groups, including fluorophosphonates and carbamates, that show little cross-reactivity with other enzyme classes (Figure 2A).[16−18] Carbamates are a privileged scaffold in this respect because their selectivity among members of the serine hydrolases can be tuned in two ways, either by modulation of the carbonyl electrophilicity

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[a]Abbreviations: CB, cannabinoid receptor; AEA, anandamide; 2-AG, 2-arachidonoylglycerol; FAAH, fatty acid amide hydrolases; MAGL, monoacylglycerol lipase; ABPP, activity-based protein profiling; ABHD6, α/β hydrolases domain-containing 6; NTE, neuropathy target esterase; TRP, transient receptor potential.

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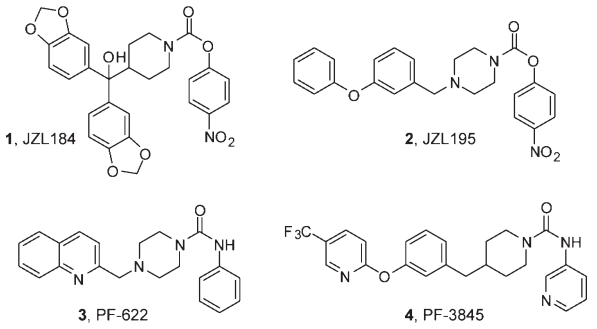


Figure 1. Structures of the MAGL-selective inhibitor **1**, the dual FAAH-MAGL inhibitor **2**, and the FAAH-selective inhibitors **3** and **4**.

(reactivity) or by modification of substituents distal to the reactive carbonyl (binding). A second attribute of serine hydrolases is that their activity state can be concurrently profiled in complex biological samples using the functional proteomic method activity-based protein profiling (ABPP).[16,19,20] ABPP of serine hydrolases is typically performed using fluorophore- or biotin-conjugated fluorophosphonate probes [e.g., FP-rhodamine (FP-Rh) or FP-biotin] that covalently label serine hydrolases, which are then resolved by SDS−PAGE and detected by in-gel fluorescence scanning (for FP-Rh) or enriched by avidin chromatography and identified by liquid chromatography−mass spectrometry (for FP-biotin).[21,22] When performed in a competitive mode, where inhibitors are preincubated with cell or tissue proteomes prior to addition of FP probes, ABPP provides a highly versatile screen for serine hydrolase inhibitors.[7,23−26] Competitive ABPP has the important advantage of testing inhibitors against numerous serine hydrolases in parallel (i.e., all of the FP-reactive enzymes that are expressed in a given cell or tissue) and, therefore, offers a powerful way to concurrently optimize the potency and selectivity of inhibitors directly in native proteomes (Figure 2B).

In our initial screen of a diverse carbamate library against the mouse brain membrane proteome, we identified two compounds (**5** and **6**) that showed good potency and promising selectivity for MAGL over the other serine hydrolases (Figure 2C,D and Table 1). At higher concentrations, both compounds also inhibited FAAH and a second brain 2-AG hydrolase, ABHD6.[6] Compounds **5** and **6** were structurally similar, each containing a six-membered ring in the form of a piperidine or piperazine, an activated *N*-4-nitrophenoxy carbamate, and a bisarylcarbinol moiety distal to the electrophilic carbonyl. However, despite their 27- and 68-fold greater activity for MAGL over FAAH in brain proteomes (Table 1), compounds **5** and **6** were not sufficiently selective for in vivo studies, since their administration to mice at doses that reduced MAGL activity also significantly blocked FAAH activity (data not shown). We therefore undertook an effort to improve the selectivity of piperazine/piperidine carbamates for inhibiting MAGL.

Structure−Activity Relationships of MAGL-Selective Inhibitors. We first asked whether the six-membered piperazine/piperidine ring was required for activity. Carbamates **9a** and **9b**, which maintained the *N*,*N*-dialkyl 4-nitrophenoxy carbamate moiety but had unbranched alkyl linkers to a bisphenyl substituted terminal carbon, and carbamate **15**, in which the piperidine was contracted into a pyrrolidine ring, were synthesized as outlined in Schemes 1 and 2. Carbamoylation

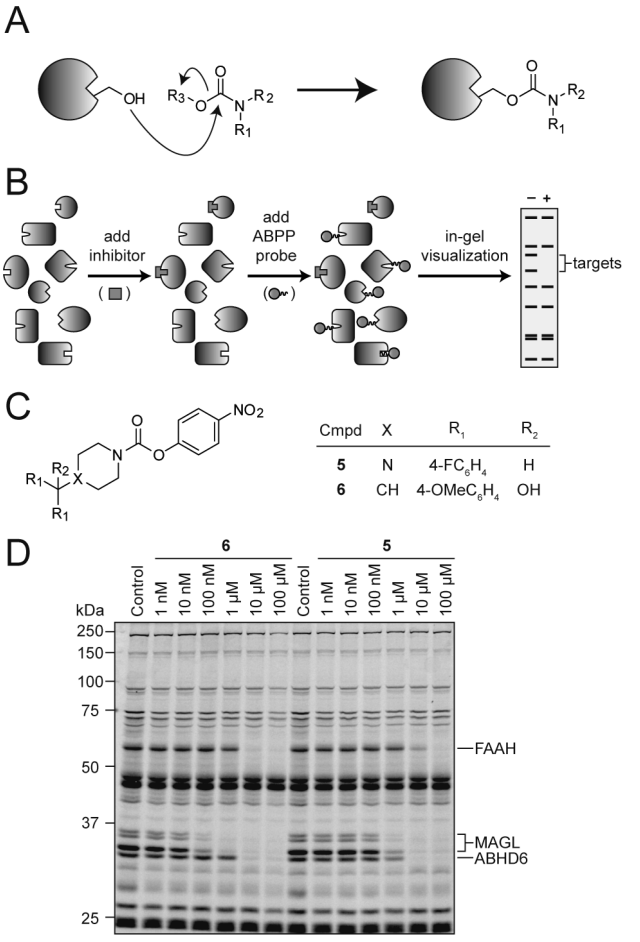


Figure 2. Competitive activity-based protein profiling (ABPP) for discovering lead carbamates that selectively inhibit MAGL. (A) Carbamates irreversibly inactivate serine hydrolases (shown as gray shapes) by carbamoylation of the nucleophilic serine. (B) Schematic depiction of competitive ABPP. Native tissue proteomes containing various serine hydrolases (gray shapes) are first incubated with a carbamate inhibitor (box) and then with an ABPP probe such as fluorphosphonate conjugated rhodamine (FP-Rh, circles). Separation and visualization of FP-Rh-labeled proteins by SDS−PAGE and in-gel fluorescence scanning, respectively, reveal the disappearance of bands that represent the serine hydrolase targets of the carbamate inhibitor (fluorescent gel shown in grayscale). (C) Structures of the lead compounds **5** and **6**. (D) Competitive ABPP showing the concentration-dependent effects of **5** and **6** on serine hydrolases activities in the mouse brain membrane proteome. **5** and **6** were incubated with proteomes for 30 min at 37 °C before addition of FP-Rh. Note that MAGL migrates as several bands between 33 and 35 kDa, as reported previously.[45]

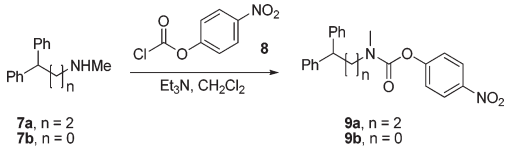
of the commercially available secondary amines **7a** and **7b** using 4-nitrophenylchloroformate (**8**) afforded **9a** and **9b**. Addition of 4-methoxyphenylmagnesium bromide (**11**) to methyl 1-benzyl-5-oxopyrrolidine-3-carboxylate (**10**) in refluxing THF gave the amide **12**, which was reduced to **13** using $LiAlH_4$. Benzyl deprotection using standard Pd/C hydrogenation conditions to afford secondary amine **14** and finally carbamoylation afforded pyrrolidine carbamate **15**. When **9a**, **9b**, and **15** were assessed by competitive ABPP against the mouse brain membrane proteome, they were found to be inactive against FAAH, MAGL, and other serine hydrolases (Table 2). These data therefore suggested that the six-membered ring in compounds **5** and **6** was required for MAGL inhibition.

Table 1. In Vitro Activity of Lead Compounds[a]

compd	FAAH	MAGL	ABHD6	selectivity
5	5410	200	900	27
6	2660	40	2110	68

[a] IC_{50} values are shown in nM unless otherwise indicated.

Scheme 1. Synthesis of Carbamates **9a** and **9b**



Scheme 2. Synthesis of Carbamate **15**

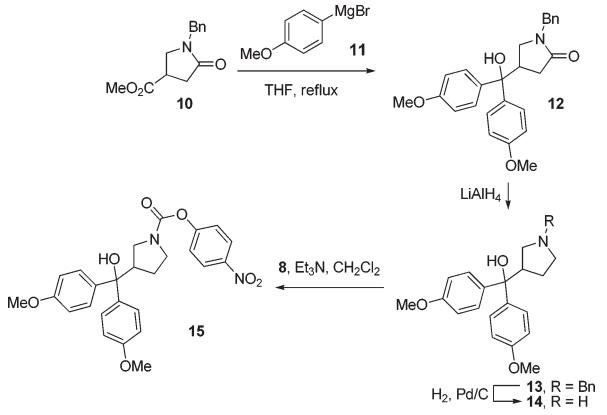
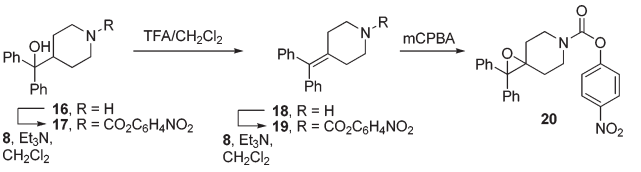


Table 2. In Vitro Activity of Carbamates Lacking a Six-Membered Ring

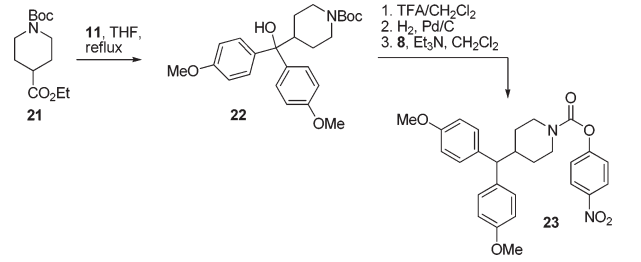
compd	FAAH	MAGL	ABHD6
9a	> 50 μM	> 50 μM	> 50 μM
9b	> 50 μM	> 50 μM	> 50 μM
15	> 50 μM	> 50 μM	> 50 μM

We next focused on making structural modifications to the portion of the molecules distal to the piperazine or piperidine ring. The synthesis of these compounds is outlined in Schemes 3−5. Carbamoylation under the usual conditions of commercially available diphenyl(piperidin-4-yl)methanol (**16**) afforded **17** (Scheme 3). Alternatively, the dibenzylic tertiary hydroxyl of **16** could be readily eliminated using TFA to afford **18**, which was then converted to the corresponding carbamate **19** using chloroformate **8**. Treatment of olefin **19** with mCPBA afforded epoxide **20** (Scheme 3). Compound **23**, containing a fully saturated bisaryl to piperidine linker, was synthesized starting from commercially available 1-*tert*-butyl 4-ethyl piperidine-1,4-dicarboxylate **21** (Scheme 4). Addition of Grignard **11** under refluxing conditions afforded Boc-protected piperidine **22**, which was then subjected to a three-step procedure involving sequential treatment with TFA/CH_2Cl_2, which concomitantly removed the Boc group and eliminated the hydroxyl, Pd/C under the usual hydrogenation conditions, which saturated the incipient olefin, and chloroformate **8**, to afford the saturated piperidine carbamate **23**. We also rigidified the bisaryl ring system by installing an ortho-oxygen bridge. This compound (**29**) was synthesized starting from xanthen-9-one **24**, which was reduced with $NaBH_4$ and chlorinated with $SOCl_2$ to afford **26**. Piperazine **28**, which was made using a two-step

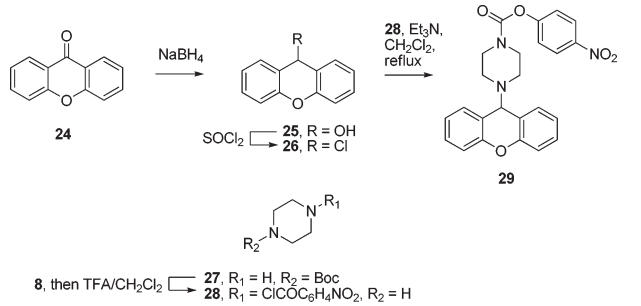
Scheme 3. Synthesis of Carbamates **17**, **19**, and **20**



Scheme 4. Synthesis of Carbamate **23**



Scheme 5. Synthesis of Carbamate **29**

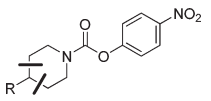


carbamoylation/deprotection sequence from Boc-piperazine, was used to alkylate **26** and afford **29**.

17, **19**, **20**, **23**, and **29** were evaluated against brain membrane proteomes using competitive ABPP. Comparison of **17** and **6** revealed that the 4-methoxy group on the distal aryl rings conferred both additional potency and selectivity for MAGL (Table 3). However, rigidification of any portion of this distal substitutent, either by installation of an olefin (**19**) or epoxide (**20**) linker between the phenyl groups and the piperidine ring or by covalently tethering the aryl rings with an oxygen bridge (**29**), reduced the activity and/or selectivity for MAGL in comparison to lead carbamates **5** and **6** (IC_{50} = 200 nM to 6 μM for **19**, **20**, and **29**, versus IC_{50} = 200 and 40 nM for **5** and **6**, respectively). **23**, in which the methanol linker of compound **17** was replaced with a methane, exhibited excellent selectivity for MAGL over FAAH (> 500-fold), despite being of slightly lower potency compared to lead compound **6** (IC_{50} = 70 nM versus 40 nM). Compound **23** provided the first evidence that it would be possible to achieve selectivity for MAGL over FAAH on the order of 300- to 1000-fold with the piperidine carbamate scaffold. We therefore continued to search for compounds that displayed this high degree of selectivity while also being more potent for MAGL than the original compound **6**.

Our next series of carbamates was motivated by the improved potency of **6** compared to **17**, which suggested that further gains in MAGL active site binding interactions could be achieved by additional substitution and/or increasing the steric bulk of the arene rings. We reasoned that such modifications might have the further benefit of increasing

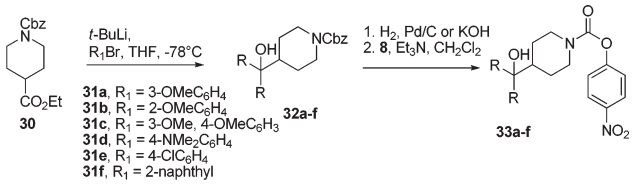
Table 3. In Vitro Activity of Carbamates Modified with Different Linkers[a]



Cmpd	R =	FAAH	MAGL	ABHD6	Selectivity
17		4340	690	1100	6
19		3500	1390	1300	3
20		5850	200	380	29
23		40080	73	550	549
29		9000	6120	2120	2

[a] IC_{50} values are shown in nM unless otherwise indicated.

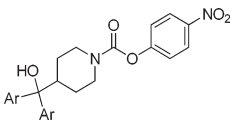
Scheme 6. Synthesis of Carbamates **33a**−**f**



the selectivity window for MAGL versus FAAH, since we hypothesized that these compounds' selectivity was in part due to the inability of the sterically encumbering bisaryl groups to fit into the relatively narrow FAAH acyl-chain binding pocket.[27] Since **6** remained at this point the most potent compound identified, we chose to maintain the piperidine carbinol scaffold while varying the aryl groups. Compounds **33a**−**f** and **1** were prepared according to Scheme 6. Addition of variously substituted aryl groups to the ester of 1-benzyl 4-ethyl piperidine-1,4-dicarboxylate (**30**) was accomplished using aryllithium anions at −78 °C, which were generated by treating the appropriate aryl bromide with *t*-BuLi at the same temperature. The resultant Cbz-protected 4-bisarylcarbinolpiperidine compounds **32a**−**f** were deprotected by hydrogenation using Pd/C (**32a**−**d**, **32f**) or aqueous KOH (**32e**) to give the crude amines, which were used directly in the carbamoylation reaction with **8** to afford the carbamates **33a**−**f**.

Evaluation of **33a** and **33b** by competitive ABPP showed that a single methoxy substituent at the 3- or 2- position of the distal aryl ring could be tolerated and that these compounds could still maintain good selectivity for MAGL (Table 4, ∼10-fold selective). Remarkably, when we incorporated two oxygen substituents, in the form of either the 3,4-dimethoxybenzene (**33c**) or 3,4-methylenedioxybenzene (**1**), we achieved excellent selectivity for MAGL (about 40- to 400-fold selective) without inhibiting any of the other serine hydrolases in mouse brain membranes as judged by our competitive ABPP gels. Compound **1**, which was ultimately advanced to in vivo pharmacological and behavioral studies,

Table 4. In Vitro Activity of Carbamates Modified at the Distal Arene Group[a]



Cmpd	Ar =	FAAH	MAGL	ABHD6	Selectivity
33a		5830	560	460	10
33b		15100	510	1100	11
33c		>50 μM	1300	6960	>38
1		4690	10	3270	426
33d		10540	2120	1020	5
33e		3360	1100	180	3
33f		4690	810	3430	6

[a] IC_{50} values are shown in nM unless otherwise indicated.

maintained a comparable MAGL-selectivity window to **23** but gained a 7-fold improvement in potency. An additional advantage was that, unlike many other compounds in the piperidine/piperazine carbamate series, **1** exhibited low activity for ABHD6, another enzyme that has been shown to hydrolyze 2-AG in vitro.[6] Further efforts to refine the aryl substitution patterns by expanding the steric bulk, by using 4-*N*,*N*-dimethylanilinebenzene (**33d**) or 2-naphthyl (**33f**) groups, or by modifying the aryl group electronics, with substituents such as 4-chlorobenzene (**33e**), did not produce more potent or more selective compounds. Since both larger (**33c**, **33d**, and **33f**) and smaller (**17**) substituents reduce the potency of these piperidine carbamates for MAGL, we attribute, in part, the high potency and selectively of **1** to be due to its unique substitution pattern and steric properties, since the methylenedioxy group exploits two points of substitution along the arene ring while maintaining a relatively small van der Waals volume.

Tuning the Piperazine Carbamate Scaffold To Create Dual FAAH-MAGL Inhibitors. Our competitive ABPP screens revealed that many compounds of the piperidine/piperazine carbamate class with activity for MAGL also tended to inhibit a select number of additional serine hydrolases in the mouse brain proteome, principally FAAH and ABHD6. This suggested that, despite their low sequence homology, MAGL, FAAH, and ABHD6 share active site similarity, a hypothesis that is reinforced by the structural relatedness of some FAAH-selective (**3** and **4**) and MAGL-selective (**1**) inhibitors, which all shared an N-electrophile tethered to a piperidine or piperazine ring motif. Since **3** and **4** derive their selectivity for FAAH by containing a relatively unreactive urea electrophile, and since **1** derives its selectivity for MAGL from the bulky piperidine bisarylcarbinol scaffold, we reasoned that an appropriate combination of **1** and **3** could potentially provide a dual FAAH-MAGL inhibitor.[28,29]

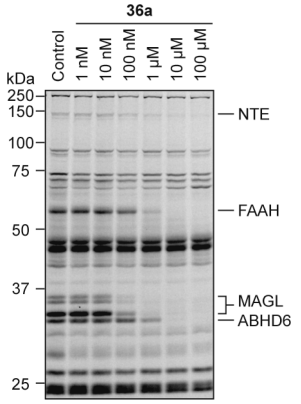
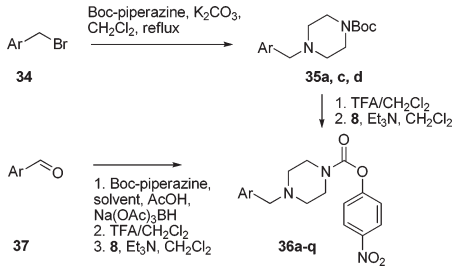


Figure 3. Concentration-dependent effects of **36a** on serine hydrolases activities in the mouse brain membrane proteome as determined by competitive ABPP. **36a** was incubated with proteomes at the indicated concentrations for 30 min at 37 °C before addition of FP-Rh probe.

Scheme 7. Synthesis of Carbamates **36a−q**

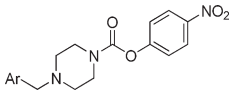


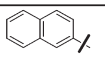
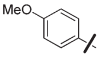
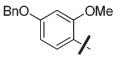
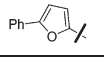
In such a compound, the FAAH-selective urea electrophile would be replaced with a carbamate to enable inactivation of MAGL (4-nitrophenoxy carbamate), and the MAGL-selective bulky bisarylcarbinol motif would be substituted with a distal group that interacted with both the FAAH and MAGL active sites.

With these general concepts in mind, we targeted the piperazine *p*-nitrophenoxy carbamate as a scaffold that was both likely to yield a dual inhibitor and also amenable to facile N-modification (**36a−q**). These compounds were synthesized by one of two routes shown in Scheme 7. Boc-piperazine was alkylated by displacement of substituted benzyl bromides using K_2CO_3 in refluxing CH_2Cl_2, and the resultant products were deprotected and carbamoylated in the usual way to afford the products. Alternatively, alkylation could be accomplished by reductive amination of Boc-piperazine with arylaldehydes using $Na(OAc)_3BH$ at room temperature.

Our initial compound in this series, the 2-napthyl substituted carbamate **36a**, showed good activity for FAAH and MAGL but also inhibited another serine hydrolase, the 150 kDa neuropathy target esterase (NTE) (Figure 3).[30,31] Since the inhibition of NTE is believed to be responsible for the delayed onset mortality induced by organophosphate reagents and also lower limb paralysis in humans,[32−34] we sought to reduce the potency of the piperazine carbamates for this target and pursued derivatives of **36a** (Table 5). While many compounds of this series fully blocked labeling of NTE at high concentrations and also inhibited NTE when administered to mice (for example, compounds **36h** at 20 mg/kg, ip; data not shown), a small subset of three compounds showed only partially blocked NTE activity at high concentrations (100 μM, Table 6). Of these compounds, **2** was most promising, since it showed the least inhibition toward NTE while maintaining excellent activity for MAGL and FAAH. Additional derivatives that also contained the *N*-3-aryloxybenzyl motif of **2**, such as **36m−q**, were also unable to fully block NTE activity at high doses (Table 7), suggesting that the poor inhibition of NTE exhibited by these compounds is

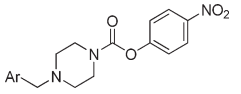
Table 5. In Vitro Activity of Initial Dual FAAH-MAGL Inhibitors[a]

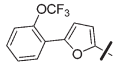
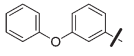


Cmpd	Ar =	FAAH	MAGL	ABHD6	NTE
36a		380	70	150	1100
36b		1040	80	490	910
36c		930	900	740	610
36d		1050	1530	1270	480
36e		4600	15	1650	4270
36f		3570	670	2860	330
36g		930	90	310	420
36h		40	50	220	80
36i		280	10	1000	890
36j		20	90	220	250

[a] IC_{50} values are shown in nM unless otherwise indicated.

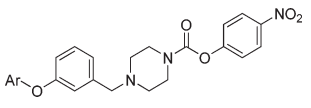
Table 6. In Vitro Activity of Dual FAAH-MAGL Inhibitors That Showed Selectivity away from NTE[a]

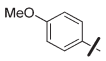
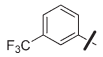


Cmpd	Ar =	FAAH	MAGL	ABHD6	NTE	% inhibition of NTE at 100 μM
36k		720	90	130	3240	73
36l		5	70	180	570	81
2		13	19	50	>50 μM	50

[a] IC_{50} values are shown in nM unless otherwise indicated.

Table 7. In Vitro Activity of Dual FAAH-MAGL Inhibitors Modified at the 3-Aryloxybenzyl Position[a]



Cmpd	Ar =	FAAH	MAGL	ABHD6	NTE	% inhibition of NTE at 100 μM
36m		40	500	1130	3000	70
36n		40	50	60	1270	73
36o		90	130	460	2900	60
36p		5	300	320	14400	57
36q		>100 μM	>100 μM	>100 μM	>100 μM	0

[a] IC_{50} values are shown in nM unless otherwise indicated.

due to the unique steric and/or electronic properties of the aryloxybenzyl substitution. That 3,5-dichlorobenzene phenyl ether **36q** was unable to inhibit any of the hydrolases in our screen indicated that there is an upper limit to the steric bulk that can be appended to the distal arene ring. Thus, while the piperidine bisarylcarbinol scaffold results in MAGL-selective inhibition, a piperazine aryloxybenzyl scaffold can be used as a scaffold for the selective dual blockade of FAAH and MAGL.

Though **2** showed weak inhibition of NTE in vitro (~50% blockade at 100 μM), no inhibition of NTE was observed following in vivo administration to mice at doses that completely blocked FAAH and MAGL (20 mg/kg ip or 100 mg/kg orally), and chronic administration of **2** to mice (20 mg/kg ip daily for 6 days) did not produce death or overt signs of discomfort.[9] That ABHD6 was also inhibited at higher concentrations suggests that **2** has the potential to function as a poly-pharmacological inhibitor for several endocannabinoid hydrolases (FAAH and two of the major brain 2-AG hydrolases, MAGL and ABHD6). The FAAH-selective inhibitor **4**, the MAGL-selective inhibitor **1**, and the dual FAAH-MAGL inhibitor **2** thus constitute a versatile arsenal of efficacious and selective pharmacological probes that can be used to interrogate the function of endocannabinoid-degrading enzymes in vivo.

Conclusions

Herein, we have described the discovery, synthesis, structure−activity relationships, and inhibitory activities of a series of piperidine/piperazine carbamates that function either as selective inhibitors of MAGL or as dual inhibitors of FAAH and MAGL in vivo. The development of compounds such as **1** and **2** underscores the utility of competitive ABPP to concurrently optimize inhibitor potency and selectivity directly in native proteomes. Indeed, considering that NTE shares little to no sequence homology with either FAAH or MAGL,[31] this common "off-target" of dual FAAH-MAGL inhibitors would likely have gone undetected without the broad and unbiased screening output provided by competitive ABPP. From a structure−activity perspective, our data suggest that the MAGL-selective inhibitors derive much of their selectivity from the piperidine ring as well as the bulky bisarylcarbinol motif distal to the electrophilic carbonyl. We have also demonstrated that by rationally changing the substitution at the 4-position of the six-membered ring, from bisarylcarbinol to 3-aryloxybenzyl, we can selectively tune the inhibitory properties of the piperidine/piperazine carbamate scaffold from being a MAGL-selective inhibitor to a dual inhibitor of MAGL and FAAH.

Since their disclosure, **1** and **2** have proven useful for selectively enhancing 2-AG signaling or concurrently augmenting both 2-AG and anandamide signaling, respectively, in a variety of in vitro and in vivo systems.[35−39] Nevertheless, we anticipate that even within this general chemical class there remains much potential for improvements in the potency and selectivity of future MAGL-selective inhibitors or dual FAAH-MAGL inhibitors. For instance, compound **36e**, while originally designed to be a dual FAAH-MAGL inhibitor, in fact displayed remarkable selectivity for MAGL over FAAH. We attribute this unique property of **36e** to the rigid and linear biphenyl group and speculate that this compound might serve as a lead for the development of next generation MAGL inhibitors with a scaffold distinct from the bisarylcarbinol of **1**. Moreover, we elected to use a 4-nitrophenoxy carbamate, since our initial structure−activity relationship efforts indicated that MAGL inhibition requires an activated leaving group. However, we hypothesize that, similar to the urea series of FAAH inhibitors,[5] hydroxypyridine leaving groups and derivatives thereof may also be sufficiently activated for MAGL inhibition and that incorporation of additional substitutions on the pyridine ring might offer a new way to tune the selectivity window of these inhibitors for MAGL versus FAAH. Recently, two crystal structures of human MAGL have been solved, one of which contained the piperazine inhibitor SAR629 bound to the catalytic serine of MAGL.[40,41] These crystal structures may help in the design of future irreversible or reversible MAGL inhibitors. Lastly, piperazine carbamates have been also reported to act as dual FAAH inhibitors/transient receptor potential V1 (TRPV1) antagonists.[42] That TRP channels can also be activated by endogenous FAAH substrates such as AEA[43] or the *N*-acyltaurines[44] suggests a high degree of homology between the binding pockets of these proteins and furthermore indicates that the piperidine/piperazine scaffold could potentially be exploited to generate polypharmacological tools that interact with MAGL, FAAH, and members of the TRP channel family.

Experimental Section

Chemistry. General Methods. All reagents were purchased from Sigma-Aldrich, Acros, Fisher, Fluka, or Maybridge and used without further purification, except where noted. Dry solvents were obtained by passing commercially available predried, oxygen-free formulations through activated alumina columns. All reactions were carried out under a nitrogen atmosphere using oven-dried glassware unless otherwise noted. Flash chromatography was performed using 230−400 mesh silica gel. NMR spectra were recorded in $CDCl_3$ on a Varian Inova-400 or a Bruker DMX-600 spectrometer and were referenced to trimethylsilane (TMS) or the residual solvent peak. Chemical shifts are reported in ppm relative to TMS, and *J* values are reported in Hz. High resolution mass spectrometry (HRMS)

experiments were performed at The Scripps Research Institute Mass Spectrometry Core on an Agilent mass spectrometer using electrospray ionization-time-of-flight (ESI-TOF). LC/MS analysis was performed on an Agilent 1100 LC/MS instrument, using a ZORBAX SB-C18, 3.5 mm, 4.6 mm × 50 mm column, a flow rate of 0.75 mL/min, detection at 220 and 254 nm, a 10−98% acetonitrile/water/0.1% formic acid gradient, and a 50−98% acetonitrile/water/0.1% formic acid gradient. The purity of compounds (≥95%) was confirmed by clean NMR spectra and elution as a single peak by LC/MS.

Compounds **1**−**6** have been described previously.[5,7,9,10]

4-Nitrophenyl 3,3-Diphenylpropyl(methyl)carbamate (9a). General Procedure A. To a stirring solution of *N*-methyl-3,3-diphenylpropan-1-amine (70 mg, 0.27 mmol) in CH_2Cl_2 (3 mL) was sequentially added triethylamine (0.1 mL, 0.7 mmol) and 4-nitrochloroformate (143 mg, 0.71 mmol). After 2 h, TLC indicated complete consumption of the starting material. The mixture was diluted with CH_2Cl_2 and poured onto saturated aqueous Na_2CO_3. The organic layer was washed thrice with 2 N NaOH, once with brine, dried over Na_2SO_4, and concentrated in vacuo. Purification of the crude oil by flash chromatography (1−2% MeOH in CH_2Cl_2) gave **9a** (52 mg, 49% yield): ^{1}H NMR ($CDCl_3$, 400 MHz) δ 8.09 (d, J = 9.2 Hz, 2H), 7.28−7.16 (m, 10H), 6.90 (d, J = 9.2 Hz, 2H), 3.78 (t, J = 7.8 Hz, 2H), 3.12 (t, J = 7.8 Hz, 2H), 2.81 (s, 3H), 2.31 (m, 2H). MS (ESI$^+$) m/z 391 $[M + H]^+$. HRMS calculated for $C_{23}H_{23}N_2O_4$ $[M + H]^+$ 391.1652, found 391.1650.

4-Nitrophenyl Benzhydryl(methyl)carbamate (9b). 9b was prepared according to general procedure A, using *N*-methyl-1, 1-diphenylmethanamine (280 mg, 1.42 mmol), 4-nitrochloroformate (313 mg, 1.56 mmol), triethylamine (1 mL, 7 mmol), and CH_2Cl_2 (10 mL). Purification of the crude oil by flash chromatography (12:1 Hex/EtOAc) gave **9b** (201 mg, 39% yield): ^{1}H NMR ($CDCl_3$, 400 MHz) δ 8.24 (d, J = 8.1 Hz, 2H), 7.42−7.25 (m, 12H), 6.73 (s, 1H), 2.93 (s, 3H). HRMS calculated for $C_{21}H_{19}N_2O_4$ $[M + H]^+$ 363.1339, found 363.1364.

1-Benzyl-4-(hydroxybis(4-methoxyphenyl)methyl)pyrrolidin-2-one (12). To a stirring solution of methyl 1-benzyl-5-oxopyrrolidine-3-carboxylate (494 mg, 2.12 mmol) in dry ether (10 mL) was added 4-methoxyphenylmagesium bromide (0.5 M in THF, 10 mL, 5 mmol). The mixture was heated to reflux, and TLC indicated completion consumption of the starting material after 12 h. The mixture was diluted with CH_2Cl_2 and poured onto saturated aqueous Na_2CO_3. The organic layer was washed once with brine, dried over Na_2SO_4, and concentrated in vacuo. Purification of the crude oil by flash chromatography (1:1 Hex/EtOAc) gave **12** (190 mg, 21% yield): ^{1}H NMR ($CDCl_3$, 400 MHz) δ 7.31−7.19 (m, 7H), 7.15 (d, J = 7.4 Hz, 2H), 6.81−6.64 (m, 4H), 4.43 (d, J = 14.8 Hz, 1H), 4.11 (d, J = 15.3 Hz, 1H), 3.70 (s, 6H), 3.48- 3.33 (m, 1H), 3.23 (dd, J = 10.0, 6.3 Hz, 1H), 3.08 (t, J = 9.5 Hz, 1H), 2.45 (ddd, J = 27.2, 17.5, 8.5 Hz, 2H). MS (ESI$^+$) m/z 440 $(M + Na)^+$.

(1-Benzylpyrrolidin-3-yl)bis(4-methoxyphenyl)methanol (13). To a −78 °C stirring solution of **12** (185 mg, 0.44 mmol) in dry ether/CH_2Cl_2 (4:1 v/v, 25 mL total) was added $LiAlH_4$ (4 M in ether, 0.5 mL, 2 mmol). The dry ice bath was removed, and the mixture was heated to reflux. After 2 h, TLC indicated complete consumption of the starting material. The mixture was diluted with CH_2Cl_2 and poured onto water. The organic layer was washed once with brine, dried over Na_2SO_4, and concentrated in vacuo. The crude oil was passed through a pad of silica to afford **13** (160 mg, 90% yield): ^{1}H NMR ($CDCl_3$, 400 MHz) δ 7.30−7.15 (m, 9H), 6.78−6.52 (m, 4H), 3.65 (d, J = 5.2 Hz, 6H), 3.52 (d, J = 12.8 Hz, 1H), 3.41 (d, J = 12.9 Hz, 1H), 3.16−3.08 (m, 1H), 2.95−2.87 (m, 1H), 2.77 (d, J = 9.5 Hz, 1H), 2.16 (dd, J = 9.5, 6.5 Hz, 1H), 2.03 (q, J = 8.9 Hz, 1H), 1.91−1.74 (m, 2H). MS (ESI$^+$) m/z 404 $[M + H]^+$.

4-Nitrophenyl 3-(Hydroxybis(4-methoxyphenyl)methyl)pyrrolidine-1-carboxylate (15). General Procedure B. To a stirring solution of the **13** (90 mg, 0.22 mmol) in EtOH (5 mL) was added 10% Pd/C (20 mg), and H_2 gas was bubbled through the mixture. After 4 h, TLC indicated complete consumption of the starting material. The mixture was diluted with CH_2Cl_2, filtered over a pad of Celite, and concentrated in vacuo. The crude was taken up in CH_2Cl_2 (10 mL), and triethylamine (1 mL, 7 mmol) and 4-nitrochloroformate (80 mg, 0.4 mmol) were sequentially added. After 2 h, TLC indicated complete consumption of the starting material. The mixture was diluted with CH_2Cl_2 and poured onto saturated aqueous Na_2CO_3. The organic layer was washed thrice with 2 N aqueous NaOH, once with brine, dried over Na_2SO_4, and concentrated in vacuo. Purification of the crude oil by flash chromatography (6:1 Hex/EtOAc) gave the **15** (30 mg, 28% yield over two steps): ^{1}H NMR ($CDCl_3$, 400 MHz) δ 8.28−8.17 (m, 2H), 7.40−7.22 (m, 6H), 6.85 (dd, J = 12.5, 5.8 Hz, 4H), 3.78 (dd, J = 3.2, 1.3 Hz, 6H), 3.71−3.30 (m, 4H), 2.34 (d, J = 14.1 Hz, 1H), 2.01−1.89 (m, 2H). HRMS calculated for $C_{26}H_{26}N_2NaO_7$ $[M + Na]^+$ 501.1632, found 501.1681.

4-Nitrophenyl 4-(Hydroxydiphenylmethyl)piperidine-1-carboxylate (17). 17 was prepared according to general procedure A, using 4-benzhydrylpiperidine (192 mg, 0.72 mmol), 4-nitrochloroformate (230 mg, 1.1 mmol), triethylamine (0.2 mL, 1.4 mmol), and CH_2Cl_2 (10 mL). Purification of the crude oil by flash chromatography (2−3% MeOH in CH_2Cl_2) gave **17** (100 mg, 32% yield): ^{1}H NMR ($CDCl_3$, 400 MHz) δ 8.22 (d, J = 9.1 Hz, 2H), 7.47 (d, J = 8.0 Hz, 4H), 7.35−7.22 (m, 8H), 4.30 (bs, 2H), 3.02 (t, J = 12.5 Hz, 1H), 2.89 (t, J = 12.2 Hz, 1H), 2.64 (m, 1H), 1.64−1.60 (m, 2H), 1.47 (m, 2H). HRMS calculated for $C_{25}H_{24}N_2NaO_5$ $[M + Na]^+$ 455.1577, found 455.1586.

4-(Diphenylmethylene)piperidine (18). To a stirring solution of 4-benzhydrylpiperidine (1.77 g, 6.7 mmol) in CH_2Cl_2 (15 mL) was added TFA (5 mL). After 7 h, TLC indicated complete consumption of the starting material. The mixture was concentrated in vacuo and then diluted with CH_2Cl_2 and saturated aqueous Na_2CO_3. The aqueous layer was extracted twice with CH_2Cl_2, and the combined organic layers were dried over Na_2SO_4 and concentrated in vacuo. The crude was passed through a pad of silica (5% MeOH in CH_2Cl_2) to give **18** (1.6 g, 96% yield): ^{1}H NMR ($CDCl_3$, 400 MHz) δ 7.34−7.23 (m, 6H), 7.11−7.08 (m, 4H), 3.24 (bs, 4H), 2.64 (m, 4H). MS (ESI$^+$) m/z 250 $[M + H]^+$.

4-Nitrophenyl 4-(Diphenylmethylene)piperidine-1-carboxylate (19). 19 was prepared according to general procedure A, using **18** (182 mg, 0.73 mmol), 4-nitrochloroformate (142 mg, 0.71 mmol), triethylamine (0.3 mL, 2.1 mmol), and CH_2Cl_2 (10 mL). The crude product was passed through a pad of silica (CH_2Cl_2) to afford **19** (281 mg, 93% yield): ^{1}H NMR ($CDCl_3$, 400 MHz) δ 8.23 (d, J = 9.3 Hz, 2H), 7.36−7.09 (m, 12H), 3.69 (m, 2H), 3.61 (m, 2H), 2.47 (m, 4H). HRMS calculated for $C_{25}H_{23}N_2O_4$ $[M + H]^+$ 415.1652, found 415.1676.

4-Nitrophenyl 2,2-Diphenyl-1-oxa-6-azaspiro[2.5]octane-6-carboxylate (20). To a stirring solution of **19** (23 mg, 0.056 mmol) in CH_2Cl_2 (5 mL) was added mCPBA (< 72%, 49 mg, < 0.2 mmol). After 2 h, TLC indicated complete consumption of the starting material, and a saturated aqueous solution of $Na_2S_2O_3$ (1 mL) was added to the reaction. After being stirred for 30 min, the mixture was diluted with EtOAc and the organic layer was washed twice with water, once with saturated aqueous Na_2CO_3, once with brine, dried over Na_2SO_4, and concentrated in vacuo. The crude was passed through a pad of silica to afford **20** (15 mg, 63% yield): ^{1}H NMR ($CDCl_3$, 400 MHz) δ 8.24 (d, J = 9.2 Hz, 2H), 7.47 (d, J = 7.6 Hz, 2H), 7.39 −7.22 (m, 8H), 4.03 (m, 2H), 3.51 (t, J = 11 Hz, 1H), 3.39 (t, J = 11 Hz, 1H), 1.87 (m, 2H), 1.38 (d, J = 13.8 Hz, 1H). HRMS calculated for $C_{25}H_{23}N_2O_5$ $[M + H]^+$ 431.1602, found 431.1626.

***tert*-Butyl 4-(Hydroxybis(4-methoxyphenyl)methyl)piperidine-1-carboxylate (22).** To a 0 °C stirring solution of 1-*tert*-butyl 4-ethyl piperidine-1,4-dicarboxylate (1.16 g, 4.5 mmol) in dry THF (10 mL) was added 4-methoxyphenylmagnesium bromide (26 mL, 13 mmol) dropwise. After the reagent addition was complete, the ice bath was removed and the mixture was heated

to 60 °C. TLC indicated complete consumption of the starting material after 9 h. The mixture was diluted with EtOAc and poured onto water. The organic layer was washed once with water and once with brine, dried over Na_2SO_4, and concentrated in vacuo. Purification of the crude oil by flash chromatography (12:1 and then 3:1 Hex/EtOAc) gave **22** (1.6 g, 82% yield): ^{1}H NMR ($CDCl_3$, 400 MHz) δ 6.82 (d, J = 8.8 Hz, 2H), 7.34 (d, J = 8.8 Hz, 2H), 4.11 (s, 2H), 3.75 (s, 6H), 2.68 (t, J = 11.7 Hz, 2H), 2.44 (dd, J = 13.2, 10.5 Hz, 1H), 2.16 (s, 1H), 1.60−1.45 (m, 2H), 1.42 (s, 9H), 1.33−1.19 (m, 2H). MS (ESI$^+$) m/z 410 (M − H_2O + H)$^+$.

4-Nitrophenyl 4-(Hydroxybis(4-methoxyphenyl)methyl)piperidine-1-carboxylate (23). To a stirring solution of **22** (90 mg, 0.21 mmol) in CH_2Cl_2 (2 mL) was added TFA (2 mL). The mixture turned red, and vigorous bubbling ensued. After 1 h, TLC indicated complete consumption of the starting material. Saturated aqueous Na_2CO_3 was added until the reaction was completely neutralized, and the aqueous layer was extracted thrice with CH_2Cl_2. The combined organic layers were washed once with brine, dried over Na_2SO_4, and concentrated in vacuo. The crude was taken up in EtOH (3 mL), and 10% Pd/C (100 mg) was added. After 12 h, the mixture was diluted with CH_2Cl_2, filtered over a pad of Celite, and concentrated in vacuo. The crude was taken up in CH_2Cl_2 (5 mL), and triethylamine (0.15 mL, 1.1 mmol) and 4-nitrochloroformate (66 mg, 0.33 mmol) were sequentially added. After 2 h, TLC indicated complete consumption of the starting material. The mixture was diluted with CH_2Cl_2 and poured onto saturated aqueous Na_2CO_3. The organic layer was washed once with brine, dried over Na_2SO_4, and concentrated in vacuo. Purification of the crude oil by flash chromatography (12:1 and then 3:1 Hex/EtOAc) gave **23** (16 mg, 15% yield over three steps): ^{1}H NMR ($CDCl_3$, 400 MHz) δ 7.30 (d, J = 9.2 Hz, 2H), 8.25 (d, J = 9.2 Hz, 2H), 7.09−6.99 (m, 4H), 6.90−6.78 (m, 4H), 3.80 (s, 6H), 3.68 (m, 2H), 3.61 (m, 2H), 2.52−2.43 (m, 4H). HRMS calculated for $C_{27}H_{29}N_2O_6$ [M + H]$^+$ 477.2020, found 477.2054.

4-Nitrophenyl 4-(9*H*-Xanthen-9-yl)piperazine-1-carboxylate (29). To a stirring solution of *N*-Boc piperazine (100 mg, 0.54 mmol) in CH_2Cl_2 (5 mL) was sequentially added 4-nitrophenylchloroformate (109 mg, 0.54 mmol) and Et_3N (75 μL, 0.54 mmol) at room temperature. After 3 h, the mixture was concentrated in vacuo. Purification of the crude mixture via flash chromatography (3:1 Hex/EtOAc) gave **27** (160 mg, 83% yield): ^{1}H NMR ($CDCl_3$, 400 MHz) δ 8.24 (d, J = 9 Hz, 2H), 7.31 (d, J = 9 Hz, 2H), 3.67 (bs, 2H), 3.54 (bs, 6H), 1.50 (s, 9H). To the intermediate (100 mg, 0.28 mmol) was added 1:1 v/v TFA/CH_2Cl_2 (2 mL) at room temperature. After 2 h, the mixture was concentrated in vacuo to afford crude **28**, which was used without further purification. To a stirring solution of 9*H*-xanthene-9-one (98 mg, 0.5 mmol) in EtOH (5 mL) was added $NaBH_4$ (38 mg, 1 mmol) at room temperature. After 4 h, the mixture was poured onto water (10 ml), stirred for 1 h, and then the product was filtered off, dried, and used directly in the next step. To the crude **25** in CH_2Cl_2 (5 mL) was added sulfurous dichloride (52 μL, 0.6 mmol) and Et_3N (84 μL, 0.6 mmol) dropwise at room temperature. After 2 h, the mixture was concentrated in vacuo to afford crude **26**. Crude **26** was redissolved in CH_3CN (10 mL), and to it was sequentially added crude **28** and Et_3N (70 μL, 0.5 mmol). After the reagent addition was complete, the mixture was heated to reflux overnight. The next morning, the mixture was concentrated in vacuo. Purification of the crude mixture via flash chromatography (1:1 Hex/EtOAc) gave **29** (201 mg, 84% yield over three steps): ^{1}H NMR ($CDCl_3$, 600 MHz) δ 8.20 (d, J = 9 Hz, 2H), 7.35 (m, 4H), 7.26 (m, 4H), 6.84 (d, J = 9 Hz, 2H), 4.90 (s, 1H), 3.57 (d, J = 27 Hz, 4H), 2.44 (d, J = 27 Hz, 4H). HRMS calculated for $C_{24}H_{21}N_3O_5$ [M + H]$^+$ 432.1554, found 432.1559.

Benzyl 4-(Hydroxybis(3-methoxyphenyl)methyl)piperidine-1-carboxylate (32a). General Procedure C. To a −78 °C stirring solution of 1-bromo-3-methoxybenzene (980 mg, 5.2 mmol) in dry THF (10 mL) was added *t*-BuLi (1.7 M in pentane, 3 mL, 5.1 mmol) dropwise. After 30 min, a solution of **30** (300 mg, 1.03 mmol) in dry THF (3 mL) was added to the reaction. After the mixture was stirred for 4 h at the same temperature, TLC indicated complete consumption of the starting material. The mixture was diluted with CH_2Cl_2 and poured onto water. The organic layer was washed twice with water, once with brine, dried over Na_2SO_4, and concentrated in vacuo. Purification of the crude oil by flash chromatography (10:1 and then 4:1 Hex/EtOAc) gave **32a** (290 mg, 61% yield): ^{1}H NMR ($CDCl_3$, 400 MHz) δ 7.37−7.26 (m, 5H), 7.20 (t, J = 8.0 Hz, 2H), 7.09−6.99 (m, 4H), 6.71 (ddd, J = 8.2, 2.5, 0.7 Hz, 2H), 5.06 (s, 2H), 4.20 (bs, 2H), 3.75 (s, 6H), 2.77 (bs, 2H), 2.61−2.43 (m, 1H), 2.31 (s, 1H), 1.51 (s, 1H), 1.34 (qd, J = 12.6, 4.4 Hz, 2H). MS (ESI$^+$) m/z 444 (M − H_2O + H)$^+$.

Benzyl 4-(Hydroxybis(2-methoxyphenyl)methyl)piperidine-1-carboxylate (32b). 32b was prepared according to general procedure C, using 1-bromo-2-methoxybenzene (980 mg, 5.2 mmol), 1-benzyl 4-ethyl piperidine-1,4-dicarboxylate (280 mg, 0.96 mmol), *t*-BuLi (1.7 M in pentane, 3 mL, 5.1 mmol), and dry THF (20 mL). Purification of the crude oil by flash chromatography (10:1 and then 4:1 Hex/EtOAc) gave **32b** (210 mg, 49% yield): ^{1}H NMR ($CDCl_3$, 400 MHz) δ 7.66 (d, J = 7.1 Hz, 2H), 7.41−7.26 (m, 5H), 7.20 (ddd, J = 8.1, 7.4, 1.7 Hz, 2H), 7.02 (td, J = 7.7, 1.1 Hz, 2H), 6.78 (dd, J = 8.2, 1.0 Hz, 2H), 5.14 (s, 2H), 4.24 (bs, 2H), 3.51 (s, 6H), 3.03 (t, J = 10.8 Hz, 1H), 2.85 (bs, 2H), 1.74−1.40 (m, 4H). MS (ESI$^+$) m/z 444 (M − H_2O + H)$^+$.

Benzyl 4-(Bis(3,4-dimethoxyphenyl)(hydroxy)methyl)piperidine-1-carboxylate (32c). 32c was prepared according to general procedure C, using 4-bromo-1,2-dimethoxybenzene (485 mg, 2.2 mmol), 1-benzyl 4-ethyl piperidine-1,4-dicarboxylate (215 mg, 0.74 mmol), *t*-BuLi (1.7 M in pentane, 1.2 mL, 2.0 mmol), and dry THF (10 mL). Purification of the crude oil by flash chromatography (6:1 Hex/EtOAc) gave **32c** (50 mg, 13% yield): ^{1}H NMR ($CDCl_3$, 400 MHz) δ 7.40−7.28 (m, 5H), 7.00 (s, 2H), 6.94 (d, J = 8.4 Hz, 2H), 6.80 (d, J = 8.4 Hz, 2H), 5.10 (s, 2H), 4.23 (bs, 2H), 3.85 (s, 6H), 3.84 (s, 6H), 2.78 (bs, 2H), 2.45 (t, J = 11.9 Hz, 1H), 1.70−1.47 (m, 2H), 1.38−1.15 (m, 2H). MS (ESI$^+$) m/z 444 (M + Na)$^+$.

Benzyl 4-(Bis(4-(dimethylamino)phenyl)(hydroxy)methyl)-piperidine-1-carboxylate (32d). 32d was prepared according to general procedure C, using 4-bromo-*N*,*N*-dimethylaniline (890 mg, 4.5 mmol), 1-benzyl 4-ethyl piperidine-1,4-dicarboxylate (220 mg, 0.75 mmol), *t*-BuLi (1.7 M in pentane, 2.5 mL, 4.3 mmol), and dry THF (20 mL). Purification of the crude oil by flash chromatography (4:1 Hex/EtOAc) gave **32d** (210 mg, 57% yield): ^{1}H NMR ($CDCl_3$, 400 MHz) δ 7.39−7.19 (m, 9H), 6.65 (d, J = 8.8 Hz, 4H), 5.08 (s, 2H), 4.20 (bs, 2H), 2.90 (s, 12H), 2.81−2.71 (m, 2H), 2.45 (dd, J = 13.3, 10.4 Hz, 1H), 1.63 (bs, 2H), 1.34−1.18 (m, 2H). MS (ESI$^+$) m/z 470 (M − H_2O + H)$^+$.

Benzyl 4-(Bis(4-chlorophenyl)(hydroxy)methyl)piperidine-1-carboxylate (32e). 32e was prepared according to general procedure C, using 1-bromo-4-chlorobenzene (920 mg, 4.8 mmol), 1-benzyl 4-ethyl piperidine-1,4-dicarboxylate (220 mg, 0.75 mmol), *t*-BuLi (1.7 M in pentane, 2.7 mL, 4.6 mmol), and dry ether (15 mL). Purification of the crude oil by flash chromatography (6:1 Hex/EtOAc) gave **32e** (220 mg, 63% yield): ^{1}H NMR ($CDCl_3$, 400 MHz) δ 7.46−7.18 (m, 13H), 5.01 (d, J = 9.2 Hz, 2H), 4.19 (bs, 2H), 2.80−2.64 (m, 2H), 2.55−2.38 (m, 1H), 1.54−1.20 (m, 4H). MS (ESI$^+$) m/z 452 (M − H_2O + H)$^+$.

Benzyl 4-(Hydroxydinaphthalen-2-ylmethyl)piperidine-1-carboxylate (32f). 32f was prepared according to general procedure C, using 2-bromonaphthalene (300 mg, 1.45 mmol), 1-benzyl 4-ethyl piperidine-1,4-dicarboxylate (106 mg, 0.36 mmol), *t*-BuLi (1.7 M in pentane, 0.6 mL, 1.1 mmol), and dry THF (6 mL). Purification of the crude oil by flash chromatography (4:1 Hex/EtOAc) gave **32f** (79 mg, 44% yield): ^{1}H NMR ($CDCl_3$, 400 MHz) δ 8.04 (m, 2H), 7.74−7.84 (m, 6H), 7.56−7.43 (m, 6H), 7.31−7.25 (m, 5H), 5.08 (s, 2H), 4.24 (bs, 2H), 2.98−2.81 (m, 3H), 1.61−1.39 (m, 2H), 1.34−1.20 (m, 2H). MS (ESI$^+$) m/z 524 (M + Na)$^+$.

4-Nitrophenyl 4-(Hydroxybis(3-methoxyphenyl)methyl)piperidine-1-carboxylate (33a). 33a was prepared according to general procedure B, using **32a** (290 mg, 0.63 mmol), 10% Pd/C (70 mg), EtOH (4 mL) in the first step and CH_2Cl_2 (10 mL), triethylamine (1 mL, 7 mmol), and 4-nitrochloroformate (150 mg, 0.75 mmol) in the second step. Purification of the crude oil by flash chromatography (5:1 and then 2:1 Hex/EtOAc) gave the product (220 mg, 73% yield over two steps): ^{1}H NMR ($CDCl_3$, 400 MHz) δ 8.22 (d, J = 9.1 Hz, 2H), 7.33−7.18 (m, 4H), 7.11−7.00 (m, 4H), 6.75 (dd, J = 8.2, 2.4 Hz, 2H), 4.29 (m, 2H), 3.79 (s, 6H), 3.01 (t, J = 12.0 Hz, 1H), 2.88 (t, J = 12.2 Hz, 1H), 2.59 (tt, J = 11.9, 3.3 Hz, 1H), 2.22−2.12 (m, 1H), 1.69−1.55 (m, 2H), 1.53−1.40 (m, 2H). HRMS calculated for $C_{27}H_{28}N_2NaO_7$ $[M + Na]^+$ 515.1789, found 515.1829.

4-Nitrophenyl 4-(Hydroxybis(2-methoxyphenyl)methyl)piperidine-1-carboxylate (33b). 33b was prepared according to general procedure B, using **32b** (210 mg, 0.47 mmol), 10% Pd/C (50 mg), EtOH (5 mL) in the first step and CH_2Cl_2 (10 mL) and triethylamine (1 mL, 7 mmol) and 4-nitrochloroformate (170 mg, 0.85 mmol) in the second step. Purification of the crude oil by flash chromatography (8:1 and then 2:1 Hex/EtOAc) gave the product (150 mg, 66% yield over two steps): ^{1}H NMR ($CDCl_3$, 400 MHz) δ 8.24 (d, J = 9.3 Hz, 2H), 7.65 (d, J = 6.0 Hz, 2H), 7.29 (d, J = 9.3 Hz, 2H), 7.21 (ddd, J = 8.1, 7.4, 1.7 Hz, 2H), 7.08- 6.95 (m, 2H), 6.80 (dd, J = 8.2, 0.9 Hz, 2H), 4.28 (t, J = 10.8 Hz, 2H), 3.54 (s, 6H), 3.13- 2.98 (m, 2H), 2.98- 2.85 (m, 1H), 1.82−1.52 (m, 4H). HRMS calculated for $C_{27}H_{28}N_2NaO_7$ $[M + Na]^+$ 515.1789, found 515.1837.

4-Nitrophenyl 4-(Bis(3,4-dimethoxyphenyl)(hydroxy)methyl)piperidine-1-carboxylate (33c). 33c was prepared according to general procedure B, using **32c** (50 mg, 0.1 mmol), 10% Pd/C (50 mg), EtOH/CH_2Cl_2 (4:1 v/v, 10 mL total) in the first step and CH_2Cl_2 (10 mL), triethylamine (1 mL, 7 mmol), and 4-nitrochloroformate (51 mg, 0.25 mmol) in the second step. Purification of the crude oil by flash chromatography (3:1 Hex/EtOAc) gave **33c** (20 mg, 38% yield over two steps): ^{1}H NMR ($CDCl_3$, 400 MHz) δ 8.24 (d, J = 9.2 Hz, 2H), 7.35−7.19 (m, 2H), 7.02 (d, J = 1.8 Hz, 2H), 6.97 (d, J = 8.4 Hz, 2H), 6.83 (d, J = 8.4 Hz, 2H), 4.31 (bs, 1H), 3.87 (s, 6H), 3.86 (s, 6H), 3.02 (t, J = 12.4 Hz, 1H), 2.89 (t, J = 13.0 Hz, 1H), 2.54 (t, J = 11.7 Hz, 1H), 1.79−1.60 (m, 2H), 1.46−1.41 (m, J = 11.9 Hz, 2H). HRMS calculated for $C_{29}H_{32}N_2NaO_9$ $[M + Na]^+$ 575.2000, found 575.2015.

4-Nitrophenyl 4-(Bis(4-(dimethylamino)phenyl)(hydroxy)methyl)piperidine-1-carboxylate (33d). 33d was prepared according to general procedure B, using **32d** (40 mg, 0.08 mmol), 10% Pd/C (50 mg), EtOH/CH_2Cl_2 (4:1 v/v, 5 mL total) in the first step and CH_2Cl_2 (8 mL), triethylamine (1 mL, 7 mmol), and 4-nitrochloroformate (40 mg, 0.2 mmol) in the second step. Purification of the crude oil by flash chromatography (6:1 Hex/EtOAc) gave **33d** (14 mg, 35% yield over two steps): ^{1}H NMR ($CDCl_3$, 400 MHz) δ 8.22 (d, J = 9.2 Hz, 2H), 7.27 (m, 6H), 6.68 (d, J = 8.9 Hz, 4H), 4.27 (bs, 2H), 3.13−2.71 (m, 14H), 2.53 (t, J = 11.9 Hz, 1H), 1.72 (d, J = 11.9 Hz, 2H), 1.39 (q, J = 12.2 Hz, 2H). HRMS calculated for $C_{29}H_{33}N_4O_4$ $[M - H_2O + H]^+$ 501.2496, found 501.2543.

4-Nitrophenyl 4-(Bis(4-chlorophenyl)(hydroxy)methyl)piperidine-1-carboxylate (33e). To a stirring solution of **32e** (90 mg, 0.19 mmol) in water/EtOH (1:3 v/v, 20 mL total) was added KOH (1.0 g, 18 mmol), and the mixture was heated to reflux. After the mixture was stirred overnight, TLC the next morning indicated complete consumption of the starting material. The mixture was diluted with CH_2Cl_2 and poured onto water. The organic layer was partitioned, and the aqueous layer was extracted thrice with CH_2Cl_2. The combined organic layers were washed once with brine, dried over Na_2SO_4, and concentrated in vacuo. The crude product was taken up in CH_2Cl_2 (10 mL), and to the solution was sequentially added triethylamine (1 mL, 7 mmol) and 4-nitrochloroformate (90 mg, 0.45 mmol). After 1 h, TLC indicated complete consumption of the starting material and the mixture was diluted with CH_2Cl_2 and poured onto water. The organic layer was washed thrice with 2 N NaOH, once with brine, dried over Na_2SO_4, and concentrated in vacuo. Purification of the crude oil by flash chromatography (2:1 Hex/EtOAc) gave **33e** (50 mg, 54% yield over two steps): ^{1}H NMR ($CDCl_3$, 400 MHz) δ 8.23 (d, J = 8.2 Hz, 2H), 7.41−7.23 (m, 10H), 4.31 (bs, 2H), 3.02 (m, 1H), 2.88 (m, 1H), 2.57 (t, J = 2.9 Hz, 1H), 1.66−1.38 (m, 4H). HRMS calculated for $C_{25}H_{22}Cl_2N_2NaO_5$ $[M + Na]^+$ 523.0798, found 523.0823.

4-Nitrophenyl 4-(Hydroxydinaphthalen-2-ylmethyl)piperidine-1-carboxylate (33f). 33f was prepared according to general procedure B, using **32f** (50 mg, 0.1 mmol), 10% Pd/C (50 mg), EtOH/CH_2Cl_2 (4:1 v/v, 10 mL total) in the first step and CH_2Cl_2 (10 mL), triethylamine (1 mL, 7 mmol), and 4-nitrochloroformate (61 mg, 0.3 mmol) in the second step. Purification of the crude oil by flash chromatography (3:1 Hex/EtOAc) gave **33f** (10 mg, 19% yield over two steps): ^{1}H NMR ($CDCl_3$, 400 MHz) δ 8.22 (d, J = 9.2 Hz, 2H), 8.06 (s, 2H), 7.85 (d, J = 7.4 Hz, 2H), 7.79 (d, J = 7.6 Hz, 4H), 7.57 (d, J = 8.7 Hz, 2H), 7.53−7.42 (m, 4H), 7.26 (dd, J = 4.9, 4.3 Hz, 2H), 4.32 (d, J = 13.5 Hz, 2H), 3.09 (t, J = 12.9 Hz, 1H), 3.03−2.85 (m, 2H), 1.75 (dd, J = 26.3, 13.0 Hz, 2H), 1.65−1.47 (m, 2H). HRMS calculated for $C_{33}H_{28}N_2NaO_5$ $[M + Na]^+$ 555.1890, found 555.1916.

***tert*-Butyl 4-(Naphthalen-2-ylmethyl)piperazine-1-carboxylate (35a). General Procedure D.** To a stirring solution of *N*-Boc-piperazine (370 mg, 2.0 mmol) in CH_2Cl_2 (20 mL) was sequentially added 2-(bromomethyl)naphthalene (350 mg, 1.6 mmol) and K_2CO_3 (1 g, 7.2 mmol). The mixture was heated to reflux. After 10 h, TLC indicated complete consumption of the starting material, so the mixture was diluted with EtOAc and poured onto water. The organic layer was washed once with brine, dried over Na_2SO_4, and concentrated in vacuo. Purification of the crude oil by flash chromatography (5:1 to 3:1 Hex/EtOAc) gave **35a** (420 mg, 76% yield): ^{1}H NMR ($CDCl_3$, 400 MHz) δ 7.82 (t, J = 7.6 Hz, 3H), 7.73 (s, 1H), 7.52−7.41 (m, 3H), 3.67 (s, 2H), 3.44 (bs, 4H), 2.43 (bs, 4H), 1.45 (s, 9H). MS (ESI^+) m/z 327 $[M + H]^+$.

***tert*-Butyl 4-Benzylpiperazine-1-carboxylate (35c). 35c** was prepared according to general procedure D, using *N*-Boc-piperazine (116 mg, 0.62 mmol), CH_2Cl_2 (6 mL), benzyl bromide (0.1 mL, 0.94 mmol), and K_2CO_3 (0.5 g, 3.6 mmol). Purification of the crude oil by flash chromatography (5:1 Hex/EtOAc) gave the **35c** (120 mg, 69% yield): ^{1}H NMR ($CDCl_3$, 400 MHz) δ 7.35−7.20 (m, 5H), 3.49 (s, 2H), 3.46−3.37 (m, 4H), 2.43−2.29 (m, 4H), 1.45 (s, 9H). MS (ESI^+) m/z 277 $[M + H]^+$.

***tert*-Butyl 4-(4-Methoxybenzyl)piperazine-1-carboxylate (35d). 35d** was prepared according to general procedure D, using *N*-Boc-piperazine (116 mg, 0.62 mmol), CH_2Cl_2 (6 mL), 1-(bromomethyl)-4-methoxybenzene (0.2 mL, 1.4 mmol), and K_2CO_3 (0.5 g, 3.6 mmol). Purification of the crude oil by flash chromatography (5:1 Hex/EtOAc) gave **35d** (134 mg, 71% yield): ^{1}H NMR ($CDCl_3$, 400 MHz) δ 7.22 (d, J = 8.6 Hz, 2H), 6.85 (d, J = 8.6 Hz, 2H), 3.79 (s, 3H), 3.45−3.40 (m, 6H), 2.43−2.31 (m, 4H), 1.45 (s, 9H). MS (ESI^+) m/z 307 $[M + H]^+$.

4-Nitrophenyl 4-(Naphthalen-2-ylmethyl)piperazine-1-carboxylate (36a). General Procedure E. Compound **35a** (420 mg, 1.1 mmol) was taken up in CH_2Cl_2/TFA (1:1 v/v, 6 mL total). After the mixture was stirred for 2 h, TLC indicated complete consumption of the starting material. The crude was concentrated in vacuo and used without purification. The intermediate was then taken up in CH_2Cl_2 (5 mL) and to the solution was added triethylamine (2 mL, 14 mmol) and 4-nitrochloroformate (300 mg, 1.5 mmol). After 3 h, TLC indicated complete consumption of the starting material. Purification of the crude oil by flash chromatography (5:1 Hex/EtOAc) gave **36a** (102 mg, 20% yield over two steps): ^{1}H NMR ($CDCl_3$, 400 MHz) δ 8.27 (d, J = 9.3 Hz, 2H), 7.88−7.82 (m, 3H), 7.78 (s, 1H), 7.62−7.43 (m, 3H), 7.36−7.30 (m, 2H), 3.76 (s, 2H), 3.72 (bs, 2H), 3.64 (bs, 2H), 2.60 (bs, 2H). HRMS calculated for $C_{22}H_{22}N_3O_4$ $[M + H]^+$ 392.1605, found 392.1638.

4-Nitrophenyl 4-(Quinolin-2-ylmethyl)piperazine-1-carboxylate (36b). General Procedure F. To a stirring solution of *N*-Boc-piperazine (268 mg, 1.44 mmol) in dry MeOH (6 mL) was sequentially added quinoline-2-carbaldehyde (175 mg, 1.1 mmol), AcOH (60 μL), and $Na(OAc)_3BH$ (475 mg, 2.2 mmol). After 15 h, TLC indicated complete consumption of the starting material. The mixture was diluted with EtOAc and poured onto saturated aqueous $NaHCO_3$. The organic layer was washed once with brine, dried over Na_2SO_4, and concentrated in vacuo. The crude oil was passed through a plug of silica, and the intermediate was taken up in CH_2Cl_2/TFA (1:1 v/v, 10 mL total). After the mixture was stirred for 2 h, TLC indicated complete consumption of the starting material. The crude was concentrated in vacuo and used without purification. The intermediate was then taken up in CH_2Cl_2 (5 mL), and to the solution was added triethylamine (1 mL, 7 mmol) and 4-nitrochloroformate (220 mg, 1.1 mmol). After 3 h, TLC indicated complete consumption of the starting material. The mixture was diluted with CH_2Cl_2 and poured onto saturated aqueous Na_2CO_3. The organic layer was washed thrice with 2 N NaOH, once with brine, dried over Na_2SO_4, and concentrated in vacuo. Purification of the crude oil by flash chromatography (3:1 Hex/EtOAc) gave **36b** (87 mg, 20% yield over three steps): ^{1}H NMR ($CDCl_3$, 400 MHz) δ 8.28−8.21 (m, 2H), 8.16 (d, J = 8.5 Hz, 1H), 8.09 (d, J = 8.5 Hz, 1H), 7.82 (d, J = 8.1 Hz, 1H), 7.72 (ddd, J = 8.4, 7.0, 1.3 Hz, 1H), 7.63 (d, J = 8.5 Hz, 1H), 7.57−7.51 (m, 1H), 7.33−7.27 (m, 2H), 3.91 (s, 2H), 3.72 (bs, 2H), 3.64 (bs, 2H), 2.76−2.52 (m, 4H). HRMS calculated for $C_{21}H_{21}N_4O_4$ $[M + H]^+$ 393.1557, found 393.1590.

4-Nitrophenyl 4-Benzylpiperazine-1-carboxylate (36c). 36c was prepared according to general procedure E, using **35c** (113 mg, 0.5 mmol) and CH_2Cl_2/TFA (6 mL total) in the first step and CH_2Cl_2 (5 mL), triethylamine (2 mL, 14 mmol), and 4-nitrochloroformate (160 mg, 0.8 mmol) in the second step. Purification of the crude oil by flash chromatography (4:1 Hex/EtOAc) gave **36c** (61 mg, 69% yield): ^{1}H NMR ($CDCl_3$, 400 MHz) δ 8.24 (d, J = 9.3 Hz, 2H), 7.36−7.32 (m, 4H), 7.29 (d, J = 9.3 Hz, 3H), 3.73−3.65 (m, 2H), 3.62−3.56 (m, 4H), 2.57−2.47 (m, 4H). HRMS calculated for $C_{18}H_{20}N_3O_4$ $[M + H]^+$ 342.1448, found 342.1470.

4-Nitrophenyl 4-(4-Methoxybenzyl)piperazine-1-carboxylate (36d). 36d was prepared according to general procedure E, using **35d** (110 mg, 0.36 mmol) and CH_2Cl_2/TFA (6 mL) in the first step and CH_2Cl_2 (5 mL), triethylamine (2 mL, 14 mmol), and 4-nitrochloroformate (160 mg, 0.8 mmol) in the second step. Purification of the crude oil by flash chromatography (4:1 Hex/EtOAc) gave **36d** (43 mg, 32% yield): ^{1}H NMR ($CDCl_3$, 400 MHz) δ 8.24 (d, J = 9.3 Hz, 2H), 7.29 (d, J = 9.3 Hz, 2H), 7.24 (d, J = 8.7 Hz, 2H), 6.88 (d, J = 8.7 Hz, 2H), 3.81 (s, 3H), 3.70−3.64 (m, 2H), 3.62−3.55 (m, 2H), 3.51 (s, 2H), 2.58−2.41 (m, 4H). HRMS calculated for $C_{19}H_{22}N_3O_5$ $[M + H]^+$ 372.1554, found 372.1581.

4-Nitrophenyl 4-(Biphenyl-4-ylmethyl)piperazine-1-carboxylate (36e). 36e was prepared according to general procedure F, using *N*-Boc-piperazine (302 mg, 1.6 mmol), dry CH_2Cl_2 (10 mL), biphenyl-4-carbaldehyde (260 mg, 1.4 mmol), AcOH (0.1 mL), and $Na(OAc)_3BH$ (593 mg, 2.8 mmol) in the first step, CH_2Cl_2/TFA (10 mL) in the second step, and CH_2Cl_2 (5 mL), triethylamine (1 mL, 7 mmol), and 4-nitrochloroformate (280 mg, 1.4 mmol) in the third step. Purification of the crude oil by flash chromatography (5:1 Hex/EtOAc) gave **36e** (300 mg, 51% yield over three steps): ^{1}H NMR ($CDCl_3$, 400 MHz) δ 8.25 (d, J = 9.2 Hz, 2H), 7.59 (dd, J = 13.0, 4.7 Hz, 4H), 7.51−7.25 (m, 7H), 3.70 (bs, 2H), 3.58 (bs, 4H), 2.65−2.36 (m, 4H). HRMS calculated for $C_{24}H_{24}N_3O_4$ $[M + H]^+$ 418.1761, found 418.1802.

4-Nitrophenyl 4-(Biphenyl-2-ylmethyl)piperazine-1-carboxylate (36f). 36f was prepared according to general procedure F, using *N*-Boc-piperazine (242 mg, 1.3 mmol), dry MeOH (6 mL), biphenyl-2-carbaldehyde (208 mg, 1.1 mmol), AcOH (60 μL), and $Na(OAc)_3BH$ (467 mg, 2.2 mmol) in the first step, CH_2Cl_2/TFA (10 mL) in the second step, and CH_2Cl_2 (5 mL), triethylamine (1 mL, 7 mmol), and 4-nitrochloroformate (220 mg, 1.1 mmol) in the third step. Purification of the crude oil by flash chromatography (5:1 Hex/EtOAc) gave **36f** (90 mg, 20% yield over three steps): ^{1}H NMR ($CDCl_3$, 400 MHz) δ 8.16 (d, J = 9.2 Hz, 2H), 8.08 (d, J = 9.1 Hz, 2H), 7.39−7.16 (m, 7H), 6.79 (d, J = 9.2 Hz, 2H), 3.54 (bs, 2H), 3.44 (bs, 4H), 2.34 (bs, 4H). HRMS calculated for $C_{24}H_{24}N_3O_4$ $[M + H]^+$ 418.1761, found 418.1800.

4-Nitrophenyl 4-(4-(Benzyloxy)-2-methoxybenzyl)piperazine-1-carboxylate (36g). 36g was prepared according to general procedure F, using *N*-Boc-piperazine (345 mg, 1.9 mmol), dry MeOH (6 mL), 4-(benzyloxy)-3-methoxybenzaldehyde (364 mg, 1.5 mmol), AcOH (60 μL), and $Na(OAc)_3BH$ (575 mg, 2.7 mmol) in the first step, CH_2Cl_2/TFA (10 mL) in the second step, and CH_2Cl_2 (5 mL), triethylamine (1 mL, 7 mmol), and 4-nitrochloroformate (300 mg, 1.5 mmol) in the third step. Purification of the crude oil by flash chromatography (4:1 Hex/EtOAc) gave **36g** (46 mg, 6% yield over three steps): ^{1}H NMR ($CDCl_3$, 400 MHz) δ 8.24 (d, J = 9.1 Hz, 2H), 7.44 (d, J = 7.3 Hz, 2H), 7.37 (dd, J = 11.2, 4.2 Hz, 2H), 7.33−7.23 (m, 3H), 6.91 (d, J = 1.4 Hz, 1H), 6.83 (d, J = 8.1 Hz, 1H), 6.77 (d, J = 8.1 Hz, 1H), 5.15 (s, 2H), 3.90 (s, 3H), 3.67 (bs, 2H), 3.59 (bs, 2H), 3.48 (s, 2H), 2.49 (bs, 4H). HRMS calculated for $C_{26}H_{28}N_3O_6$ $[M + H]^+$ 478.1973, found 478.2014.

4-Nitrophenyl 4-((5-Phenylthiophen-2-yl)methyl)piperazine-1-carboxylate (36h). 36h was prepared according to general procedure F, using *N*-Boc-piperazine (373 mg, 2.0 mmol), dry THF (20 mL), 5-phenylthiophene-2-carbaldehyde (372 mg, 2.0 mmol), AcOH (0.2 mL), and $Na(OAc)_3BH$ (796 mg, 3.8 mmol) in the first step, CH_2Cl_2/TFA (10 mL) in the second step, and CH_2Cl_2 (15 mL), triethylamine (2 mL, 14 mmol), and 4-nitrochloroformate (400 mg, 2.0 mmol) in the third step. Purification of the crude oil by flash chromatography (6:1 Hex/EtOAc) gave **36h** (250 mg, 30% yield over three steps): ^{1}H NMR ($CDCl_3$, 400 MHz) δ 8.23 (d, J = 9.1 Hz, 2H), 7.58 (d, J = 7.6 Hz, 2H), 7.36 (t, J = 7.6 Hz, 2H), 7.32−7.22 (m, 3H), 7.16 (d, J = 3.6 Hz, 1H), 6.89 (d, J = 3.5 Hz, 1H), 3.77 (s, 2H), 3.71 (bs, 2H), 3.62 (bs, 2H), 2.65−2.55 (m, 4H). HRMS calculated for $C_{22}H_{22}N_3O_4S$ $[M + H]^+$ 424.1325, found 424.1347.

4-Nitrophenyl 4-((4-Phenylthiophen-2-yl)methyl)piperazine-1-carboxylate (36i). 36i was prepared according to general procedure F, using *N*-Boc-piperazine (343 mg, 1.9 mmol), dry MeOH (10 mL), 4-phenylthiophene-2-carbaldehyde (260 mg, 1.4 mmol), AcOH (0.1 mL), and $Na(OAc)_3BH$ (738 mg, 3.48 mmol) in the first step; CH_2Cl_2/TFA (10 mL) in the second step, and CH_2Cl_2 (5 mL), triethylamine (1 mL, 7 mmol), and 4-nitrochloroformate (294 mg, 1.5 mmol) in the third step. Purification of the crude oil by flash chromatography (3:1 Hex/EtOAc) gave **36i** (132 mg, 28% yield over three steps): ^{1}H NMR ($CDCl_3$, 400 MHz) δ 8.28−8.21 (m, 2H), 7.70−7.52 (m, 2H), 7.47−7.34 (m, 3H), 7.33−7.19 (m, 4H), 3.81 (d, J = 0.5 Hz, 2H), 3.72 (bs, 2H), 3.63 (bs, 2H), 2.75−2.52 (m, 4H). HRMS calculated for $C_{22}H_{22}N_3O_4S$ $[M + H]^+$ 424.1325, found 424.1345.

4-Nitrophenyl 4-((5-Phenylfuran-2-yl)methyl)piperazine-1-carboxylate (36j). 36j was prepared according to general procedure F, using *N*-Boc-piperazine (348 mg, 1.9 mmol), dry CH_2Cl_2 (10 mL), 5-phenylfuran-2-carbaldehyde (285 mg, 1.66 mmol), AcOH (1 mL), and $Na(OAc)_3BH$ (807 mg, 3.8 mmol) in the first step, CH_2Cl_2/TFA (10 mL) in the second step, and CH_2Cl_2 (5 mL), triethylamine (1 mL, 7 mmol), and 4-nitrochloroformate (372 mg, 1.85 mmol) in the third step. Purification of the crude oil by flash chromatography (3:1 Hex/EtOAc) gave **36j** (196 mg, 29% yield over three steps): ^{1}H NMR ($CDCl_3$, 400 MHz) δ 8.33−8.11 (m, 2H), 7.67 (dd, J = 7.4, 0.9 Hz, 2H), 7.46−7.32 (m, 2H), 7.27 (ddd, J = 5.9, 3.5, 0.9 Hz, 3H), 6.61 (d, J = 3.2 Hz, 1H), 6.32 (d, J = 3.3 Hz, 1H), 3.70 (s, 4H), 3.63 (s, 2H), 2.61 (s, 4H). HRMS calculated for $C_{22}H_{21}N_3NaO_5$ $[M + Na]^+$ 430.1373, found 430.1383.

4-Nitrophenyl 4-(5-Bromo-2-hydroxybenzyl)piperazine-1-carboxylate (36k). 36k was prepared according to general procedure F, using *N*-Boc-piperazine (367 mg, 2.0 mmol), dry THF (20 mL), 5-bromo-2-hydroxybenzaldehyde (366 mg, 1.8 mmol), AcOH (0.2 mL), and $Na(OAc)_3BH$ (799 mg, 3.8 mmol) in the first step, CH_2Cl_2/TFA (6 mL) in the second step, and CH_2Cl_2 (5 mL), triethylamine (2 mL, 14 mmol), and 4-nitrochloroformate (400 mg, 2.0 mmol) in the third step. Purification of the crude oil by flash chromatography (3:1 Hex/EtOAc) gave **36k** (140 mg, 16% yield over three steps): ^{1}H NMR ($CDCl_3$, 400 MHz) δ 8.27 (d, J = 9.3 Hz, 2H), 7.42−7.24 (m, 3H), 7.15 (d, J = 2.4 Hz, 1H), 6.76 (d, J = 8.6 Hz, 1H), 3.82−3.51 (m, 6H), 2.67 (s, 4H). HRMS calculated for $C_{18}H_{19}BrN_3O_5$ $[M + H]^+$ 436.0503, found 436.0528.

4-Nitrophenyl 4-((5-(2-(Trifluoromethoxy)phenyl)furan-2-yl)methyl)piperazine-1-carboxylate (36l). 36l was prepared according to general procedure F, using *N*-Boc-piperazine (427 mg, 2.3 mmol), dry MeOH (10 mL), 5-(2-(trifluoromethoxy)phenyl)furan-2-carbaldehyde (244 mg, 0.95 mmol), AcOH (0.1 mL), and $Na(OAc)_3BH$ (410 mg, 1.9 mmol) in the first step;, CH_2Cl_2/TFA (10 mL) in the second step, and CH_2Cl_2 (5 mL), triethylamine (1 mL, 7 mmol), and 4-nitrochloroformate (200 mg, 1.0 mmol) in the third step. Purification of the crude oil by flash chromatography (4:1 Hex/EtOAc) gave **36l** (110 mg, 24% yield over three steps): ^{1}H NMR ($CDCl_3$, 400 MHz) δ 8.24 (d, J = 9.3 Hz, 2H), 7.91 (ddd, J = 7.6, 1.7, 0.6 Hz, 1H), 7.52−7.15 (m, 5H), 6.84 (d, J = 3.3 Hz, 1H), 6.38 (d, J = 3.4 Hz, 1H), 3.76 (bs, 4H), 3.64 (bs, 2H), 2.63 (bs, 4H). HRMS calculated for $C_{23}H_{21}F_3N_3O_6$ $[M + H]^+$ 492.1377, found 492.1417.

4-Nitrophenyl 4-(3-(*p*-Tolyloxy)benzyl)piperazine-1-carboxylate (36m). 36m was prepared according to general procedure F, using *N*-Boc-piperazine (373 mg, 2.0 mmol), dry THF (20 mL), 3-(4-methylphenoxy)benzaldehyde (425 mg, 2.00 mmol), AcOH (0.2 mL), and $Na(OAc)_3BH$ (848 mg, 4.0 mmol) in the first step, CH_2Cl_2/TFA (15 mL) in the second step, and CH_2Cl_2 (6 mL), triethylamine (3 mL, 21 mmol), and 4-nitrochloroformate (588 mg, 2.92 mmol) in the third step. Purification of the crude oil by flash chromatography (2:1 Hex/EtOAc) gave **36m** (456 mg, 51% yield over three steps): ^{1}H NMR ($CDCl_3$, 300 MHz) δ 8.26−8.21 (m, 2H), 7.32−6.84 (m, 10H), 3.67 (m, 2H), 3.58 (m, 2H), 3.53 (s, 2H), 2.50 (m, 4H), 2.34 (s, 3H). $C_{25}H_{26}N_3O_5$ $[M + H]^+$ 448.1867, found 448.1911.

4-Nitrophenyl 4-(3-(Pyridin-2-yloxy)benzyl)piperazine-1-carboxylate (36n). 36n was prepared according to general procedure F, using *N*-Boc-piperazine (305 mg, 1.64 mmol), dry CH_2Cl_2 (10 mL), 3-(pyridin-2-yloxy)benzaldehyde (104 mg, 0.52 mmol), AcOH (0.1 mL), and $Na(OAc)_3BH$ (233 mg, 1.1 mmol) in the first step, CH_2Cl_2/TFA (5 mL) in the second step, and CH_2Cl_2 (5 mL), triethylamine (1 mL, 7 mmol), and 4-nitrochloroformate (100 mg, 0.50 mmol) in the third step. Purification of the crude oil by flash chromatography (2:1 Hex/EtOAc) gave the product (85 mg, 38% yield over three steps): ^{1}H NMR ($CDCl_3$, 400 MHz) δ 8.31−8.16 (m, 3H), 7.70 (ddd, J = 8.3, 7.2, 1.9 Hz, 1H), 7.37 (t, J = 7.7 Hz, 1H), 7.33−7.24 (m, 2H), 7.17 (d, J = 8.7 Hz, 2H), 7.09−7.03 (m, 1H), 7.01 (ddd, J = 7.2, 5.1, 0.8 Hz, 1H), 6.93 (dd, J = 8.3, 0.8 Hz, 1H), 3.68 (bs, 2H), 3.59 (bs, 4H), 2.53 (bs, 4H). HRMS calculated for $C_{23}H_{23}N_4O_5$ $[M + H]^+$ 435.1663, found 435.1699.

4-Nitrophenyl 4-(3-(4-Methoxyphenoxy)benzyl)piperazine-1-carboxylate (36o). 36o was prepared according to general procedure F, using *N*-Boc-piperazine (559 mg, 3.0 mmol), dry THF (20 mL), 3-(4-methoxyphenoxy)benzaldehyde (457 mg, 2.00 mmol), AcOH (0.2 mL), and $Na(OAc)_3BH$ (848 mg, 4.0 mmol) in the first step, CH_2Cl_2/TFA (15 mL) in the second step, and CH_2Cl_2 (6 mL), triethylamine (3 mL, 21 mmol), and 4-nitrochloroformate (653 mg, 3.24 mmol) in the third step. Purification of the crude oil by flash chromatography (2:1 Hex/EtOAc) gave **36o** (343 mg, 37% yield over three steps): ^{1}H NMR ($CDCl_3$, 300 MHz) δ 8.26−8.21 (m, 2H), 7.32−7.20 (m, 3H), 7.05−6.80 (m, 7H), 3.80 (s, 3H), 3.67 (m, 2H), 3.58 (m, 2H), 3.52 (s, 2H), 2.49 (m, 4H). $C_{25}H_{26}N_3O_6$ $[M + H]^+$ 464.1816, found 464.1882.

4-Nitrophenyl 4-(3-(3-(Trifluoromethyl)phenoxy)benzyl)piperazine-1-carboxylate (36p). 36p was prepared according to general procedure G, using *N*-Boc-piperazine (559 mg, 3.0 mmol), dry THF (20 mL), 3-(3-trifluoromethylphenoxy)benzaldehyde (532 mg, 2.00 mmol), AcOH (0.2 mL), and $Na(OAc)_3BH$ (848 mg, 4.0 mmol) in the first step, CH_2Cl_2/TFA (15 mL) in the second step, and CH_2Cl_2 (6 mL), triethylamine (3 mL, 21 mmol), and 4-nitrochloroformate (648 mg, 3.22 mmol) in the third step. Purification of the crude oil by flash chromatography (2:1 Hex/EtOAc) gave **36p** (321 mg, 32% yield over three steps): ^{1}H NMR ($CDCl_3$, 300 MHz) δ 8.26−8.21 (m, 2H), 7.50−6.90 (m, 10H), 3.68 (m, 2H), 3.59 (m, 2H), 3.56 (s, 2H), 2.51 (m, 4H). $C_{25}H_{23}F_3N_3O_5$ $[M + H]^+$ 502.1584, found 502.1639.

4-Nitrophenyl 4-(3-(3,5-Dichlorophenoxy)benzyl)piperazine-1-carboxylate (36q). 36q was prepared according to general procedure G, using *N*-Boc-piperazine (373 mg, 2.0 mmol), dry THF (20 mL), 3-(3,5-dichlorophenoxy)benzaldehyde (534 mg, 2.00 mmol), AcOH (0.2 mL), and $Na(OAc)_3BH$ (848 mg, 4.0 mmol) in the first step, CH_2Cl_2/TFA (15 mL) in the second step, and CH_2Cl_2 (6 mL), triethylamine (3 mL, 21 mmol), and 4-nitrochloroformate (798 mg, 3.96 mmol) in the third step. Purification of the crude oil by flash chromatography (3:1 Hex/EtOAc) gave **36q** (422 mg, 42% yield over three steps): ^{1}H NMR ($CDCl_3$, 300 MHz) δ 8.27−8.22 (m, 2H), 7.38−6.93 (m, 7H), 6.86 (m, 2H), 3.70 (m, 2H), 3.61 (m, 2H), 3.54 (s, 2H), 2.51 (m, 4H). $C_{24}H_{22}Cl_2N_3O_5$ $[M + H]^+$ 502.0931, found 502.1004.

Preparations of Mouse Brain Membrane Proteomes. Tissues were Dounce-homogenized in PBS, pH 7.5, followed by a low-speed spin (1400*g*, 5 min) to remove debris. The supernatant was then subjected to centrifugation (64000*g*, 45 min) to provide the cytosolic fraction in the supernatant and the membrane fraction as a pellet. The pellet was washed and resuspended in PBS buffer by sonication. Total protein concentration in each fraction was determined using a protein assay kit (Bio-Rad). Samples were stored at −80 °C until use.

Competitive ABPP Experiments. Brain membrane proteomes were diluted to 1 mg/mL in PBS in a 50 μL total reaction volume. Inhibitors were added at the indicated concentration, and the mixtures were incubated for 30 min at 37 °C. FP-rhodamine was then added at a final concentration of 1 μM. After an additional 30 min at 25 °C, the reactions were quenched with 4× SDS−PAGE loading buffer, boiled for 5 min at 90 °C, subjected to SDS−PAGE, and visualized in-gel using a flatbed fluorescence scanner (Hitachi). Concentration-dependent inhibition curves were obtained from integrated gel band intensities (ImageJ) and were fit using Prism software (GraphPad) to obtain effector concentration for half-maximal response values (IC_{50}). The IC_{50} values shown in Tables 1−6 are the mean values of at least triplicate measurements, and the standard error of the mean (SEM) is < 25% of the indicated value in all cases.

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