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ARTICLE

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Design and Discovery of a Selective Small Molecule κ Opioid Antagonist (2-Methyl-*N*-((2′-(pyrrolidin-1-ylsulfonyl)biphenyl-4-yl)-methyl)propan-1-amine, PF-4455242)

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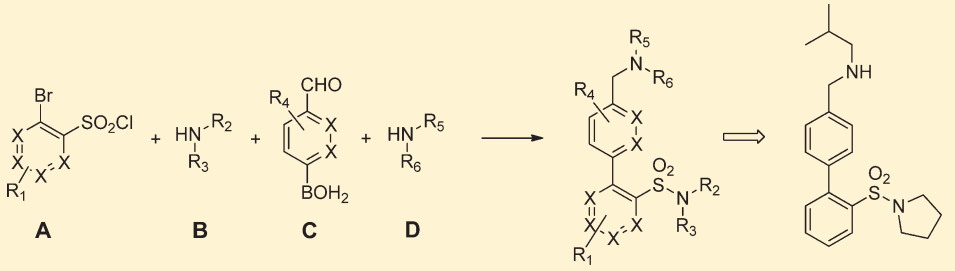
Supporting Information

ABSTRACT:



By use of parallel chemistry coupled with physicochemical property design, a series of selective κ opioid antagonists have been discovered. The parallel chemistry strategy utilized key monomer building blocks to rapidly expand the desired SAR space. The potency and selectivity of the in vitro κ antagonism were confirmed in the tail-flick analgesia model. This model was used to build an exposure−response relationship between the κ K_i and the free brain drug levels. This strategy identified 2-methyl-*N*-((2′-(pyrrolidin-1-ylsulfonyl)biphenyl-4-yl)methyl)propan-1-amine, PF-4455242, which entered phase 1 clinical testing and has demonstrated target engagement in healthy volunteers.

INTRODUCTION

The opioid family of G-protein-coupled receptors has long been a target class of interest to the medicinal chemistry community. This family consists of three subtypes μ, δ, and κ, and all have expression throughout the central nervous system.[1]

The μ opioid receptor has been the most characterized and is the target for many of the analgesic drugs on the market.[2] One of the most famous is morphine which displays effective pain control but can cause addiction, respiratory depression, and constipation. The role of the δ receptor is less clear, though it is likely to participate in processing pain signals as well.[3] In the late 1980s, the κ receptor was considered a promising target for pain, potentially lacking addiction/abuse liability. Clinical trials with spiradoline showed significant dysphoria, ending most development of agonists as potential pain therapeutics.[4]

Dynorphin is the native peptide agonist at the κ receptor, and changes in dynorphin levels in the nucleus accumbens in response to stress may be noteworthy. Most depressed patients exhibit a reduced ability to experience pleasure (anhedonia) and loss of motivation. Reward is mediated by the ventral tegmental area (VTA) nucleus accumbens (nAcc) dopaminergic pathway which is modulated (inhibited) by the κ receptors located directly on dopaminergic containing cells that project to the nAcc. Dynorphin up-regulation in the nucleus accumbens shell is stimulated by stress and various drugs of abuse and causes anhedonia-like effects potentially linking κ antagonism as a path to treating depression.[5]

Clinical evidence provides indirect support that κ receptors may be a viable target for the development of a novel antidepressant. Buprenorphine (BUP), a partial μ agonist/κ antagonist, was reported to be effective as a pharmacological treatment for affective disorders. A double blind investigation showed BUP to induce strong antidepressant effects in patients with endogenous depression.[6] Additionally, depressive symptoms were found to be significantly decreased with BUP treatment in heroin addicted patients who were depressed at intake.[7]

Preclinically it has been reported that κ blockade has afforded antidepressant activity in several animal models and is likely to dampen the decreased reward associated with excessive stimulation of κ receptors. Antidepressant activity has been reported with κ antagonists in the forced swim[8] and social defeat assays.[9]

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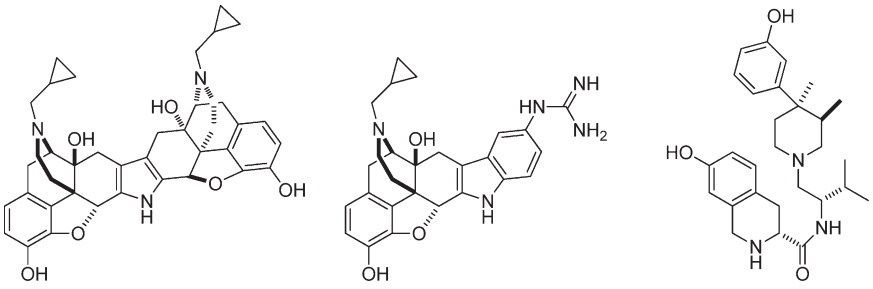


Figure 1. Structures of nor-BNI (**1**), 5′-GNTI (**2**), and JDTic (**3**).

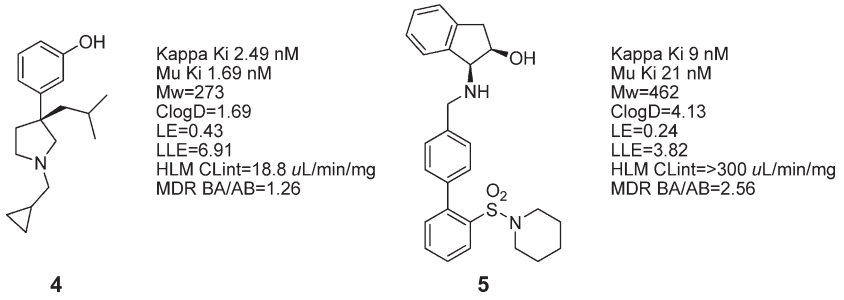


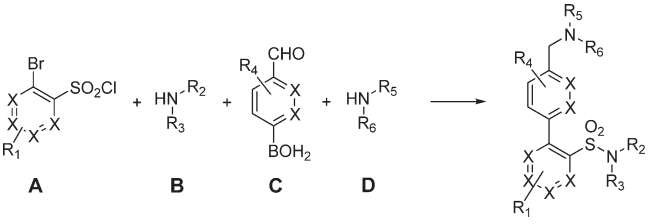
Figure 2. HTS lead structures **4** and **5**.

In addition, κ blockade has been suggested as a treatment for addiction with antagonists showing activity in reinstatement of cocaine condition place preference.[10] With the preclinical and limited clinical information, a program was engaged to identify a selective κ opioid antagonist to potentially take forward into depression and substance abuse clinical trials.

The discovery of selective, nonpeptidic κ opioid ligands has received much attention from the medicinal chemistry community. The design of κ ligands has been mainly influenced by two structural motifs, the morphinans and the 4-phenylpiperidines, which also supplied many of the breakthroughs for the discovery of selective δ and μ opioid ligands. Structural modifications of the basic morphine and naltrexone skeletons, which demonstrate affinity for all opioid receptors and modest selectivity for the μ receptor, have delivered compounds with appreciable in vitro κ receptor selectivity. Nor-BNI (**1**)[11] and 5′-GNTI (**2**)[12] (Figure 1) are two such compounds that demonstrate this principle of modulating receptor selectivity by chemical modifications of the morphinan basic skeleton. As those two molecules suggest, gains in selectivity were realized concomitant with an overall increase in molecular volume and weight, relative to the basic opioid pharmacophore (e.g., morphine or naltrexone).

Similarly, the 4-phenylpiperidines have inspired the design of κ opioid receptor selective ligands and have produced selective μ opioid receptor ligands.[13,14] Carrol and co-workers demonstrated that κ selectivity could be delivered from the phenylpiperidines by modification of the *N*-alkyl side chain.[15] JDTic (**3**) was discovered from these efforts (Figure 1).[16] Compound **3** exhibits high selectivity for the κ receptor over the μ and δ receptors and displays antagonist functional activity, thus suggesting that similar conformational aspects influenced functional activity for this class across both the μ and κ receptors. Similar to **1** and **2** the discovery of **3** suggested that selectivity could be realized with an increase in the molecular size and weight relative to the basic μ selective phenylpiperidine scaffold. The physicochemical properties and structural complexity of **1**−**3** are not in line with marketed CNS drugs.[17] This makes these templates challenging lead series to develop a safe orally bioavailable compound.

Scheme 1. Parallel Chemistry Enablement from Diverse Monomer Sets (X = CH, CR, or N)[a]

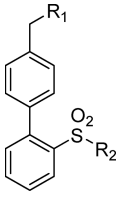


[a] Parallel synthesis reagents and conditions: (a) step 1, sulfonyl chloride **A** (1 equiv), amine **B** (1 equiv), triethylamine (2 equiv), CH_2Cl_2, 30 h, then boronic acid **C**, 1,2 dichloroethane, sodium carbonate (aq), $Pd(dppf)Cl_2$ (20%), 80 °C, 18 h, then amine **D** (1.2 equiv), sodium triacetoxyborohydride (3 equiv), 18 h. Followed by acid resin (SCX SPE) and HPLC.

RESULTS AND DISCUSSION

With this background knowledge in hand we launched a high throughput screen (HTS) to enable the discovery of novel and selective κ opioid receptor antagonist with favorable pharmaceutical and pharmacokinetic properties. The high throughput screen yielded two major classes of compounds depicted in Figure 2, as represented by the phenylpyrrolidines (e.g., compound **4**) and the biphenylamines (e.g., compound **5**). Phenylpyrrolidine **4** exhibited high affinity for the κ receptor and modest selectivity, exceptional ligand efficiency[18] (LE = 0.43),

Table 1. Biphenyl Opioid SAR and in Vitro ADME Properties[a]



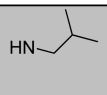
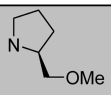
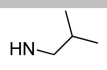
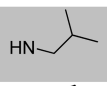
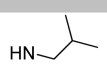
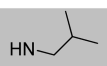
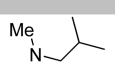
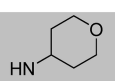
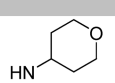
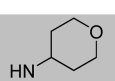
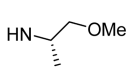
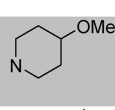
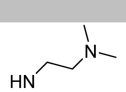
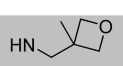
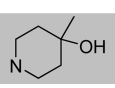
#	R1	R2	Kappa K_i	Mu K_i	HLM	MDR	ClogD
6			4.7	109	86	2.1	2.05
7			3.5	198	17.6	1.63	2.08
8			62	508	<8	1.97	2.32
9			7.5	391	58.9	1.88	2.60
10			10.0	1260	99.7	1.29	3.10
11			3.0	64	27.6	1.96	2.32
12			25	751	17.7	1.96	2.3
13			75	684	292	1.76	3.59
14			33	1007	13.4	1.78	1.68
15			14	740	15.1	2.77	1.47
16			11	70	20.4	2.14	1.7
17			8.6	433	38.7	1.99	2.16
18			2.2	89	117	1.68	2.68
19			5.8	101	20	4.68	1.06
20			2.6	193	68.9	1.72	1.87
21			13	>3000	255	1.73	1.43
22			0.5	94	60	2.62	1.98

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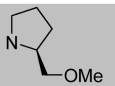
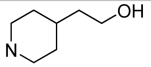
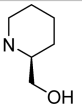
Table 1. Continued

#	R1	R2	Kappa K_i	Mu K_i	HLM	MDR	ClogD
23			1.8	66	68.2	1.98	2.45
24			3.7	207	146	2.7	2.17
25			1.1	32.0	163	14.0	1.48

[a] Human κ and μ K_i: Potency reported in nM as average of $n = 3$. HLM human liver microsomal clearance in (mL/min)/kg. MDR is the efflux ratio BA/AB. Data for compound **1** (nor-BNI): κ $K_i = 0.85$; μ $K_i = 79.9$ nM; δ $K_i = 65.2$ nM.

and lipophilic efficiency[19] (LLE = 6.91) values. The compound was characterized by extremely low molecular weight but was determined to be a functional agonist at κ in the GTP-γS assay. The biphenylamine **5** similarly exhibited high affinity for the κ receptor (9 nM) and modest selectivity over μ (2×). Importantly, compound **5** was found to be a functional antagonist at both κ and μ. On the basis of our knowledge of opioid medicinal chemistry, we anticipated that the phenylpyrrolidines **4** would be limited by a series of issues that characterize the structural motif. For example, we anticipated that the functional activity would vary with different N substitutions, and as previous literature suggested, antagonists would likely require large alkyl substitutions.[20] We also anticipated that issues such as CYP2D6 metabolism, P-gp, and HERG channel interaction would complicate the lead optimization of the series given the basic tertiary amine moiety.[21,22] We anticipated that the solution space, for example, reduction of lipophilicity and modulation the pK_a of the nitrogen, was limited because of the synthetic chemistry limitations for rapid modification of the core phenylpyrrolidine. We also anticipated that the phenol moiety, which was important for pharmacological activity, would pose a risk for secondary metabolism and that phenol surrogates would be required to optimize in vivo performance.[23] Typically phenol replacements contain acidic hydrogen bond donors and they also pose increased risk for P-glycoprotein efflux (P-gp). By applying similar prospective thinking, we rationalized that the chief issue with the biphenylamines was molecular weight and lipophilicity reduction. While we anticipated that issues such as CYP2D6 metabolism, HERG channel interaction, and P-gp efflux were likely to emerge, we rationalized that we would more efficiently probe the solution space within this core because the intrinsic synthetic enablement provided us with the option to vary polarity, hydrogen bond donor number, and pK_a readily. Consequently we investigated the rapid exploration of the lead series represented by compound **5**.

Although a potent lead was identified, compound **5** is large for a lead structure with a molecular weight of 462 and is fairly lipophilic with ClogD = 4.13. Both of these properties fall outside the ideal range for central nervous system (CNS) drugs which display an average molecular weight of 305 and an average ClogD of 1.7.[24] Even with a high degree of potency for **5**, these unoptimized physical chemical properties result in a poor ligand efficiency (LE = 0.24) and a low lipophilic ligand efficiency (LLE = 3.82). The high molecular weight and high lipophilicity significantly contribute to the undesirable in vitro ADME properties of **5**. It displays high human liver microsomal clearance with an intrinsic clearance of >300 (mL/min)/kg, suggesting a low projected human bioavailability.[25] In addition, it appears to be a P-gp substrate with an MDR flux ratio of 2.56.[26] The high P450 mediated clearance and the potential brain penetration liability result in this molecule requiring very high doses to achieve CNS exposure. The high projected doses and the poor physicochemical properties make this molecule a significant attrition risk.[27] Selectivity over μ was another key area to improve, and a selectivity of >20× was targeted to allow for 80% κ receptor occupancy with less than 10% μ receptor occupancy.[28]

While compound **5** was not the ideal lead from a binding efficiency or a physical chemical property perspective, it did have a large advantage in that it could be chemically enabled to deliver diverse analogues in a parallel chemistry format. By focusing the chemistry strategy on enabling chemistry, we could explore a variety of binding mode hypotheses and improve the physical chemical properties in a rapid fashion.[29] Toward this end, the synthetic chemistry was designed to utilize diverse building blocks that were available commercially or could be readily synthesized. Monomers A and C were prepared (Scheme 1) with a variety of substituted phenyl groups and pyridine heterocycles. In addition, there are greater than 500 primary and secondary amines in our internal file and available externally for use in library chemistry. By preparation of only five A and C monomer building blocks, the synthesis of over 6.25 million compounds was enabled.

With the extraordinary number of compounds that one could consider making, a design plan was put in place targeting improved physical chemical properties. The pyridine heterocycles were designed for monomers A and C to lower lipophilicity to improve safety and clearance with the hope of maintaining potency and improving selectivity.[30] In addition the pyridine C ring monomers would reduce the pK_a of the amine, reducing a potential HERG and P-gp liability. The large amine monomer set was reduced by considering the molecular weight of the amine building block. The biphenylsulfonyl core has a minimum molecular weight of 230 g/mol; limiting the amines to a molecular weight of less than 125 g/mol significantly reduced the potential final compounds under consideration. By use of the reduced monomer sets, the library was enumerated and the final compounds were filtered based on physical chemical properties. The decision

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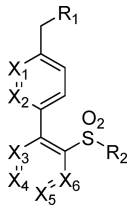
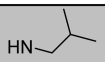
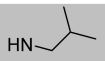
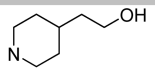
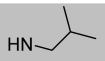
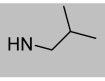
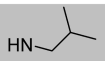
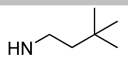
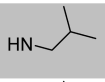
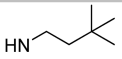
Table 2. Heteroaryl Opioid SAR and in Vitro ADME Properties[a]



#	X≠CH	R1	R2	Kappa K_i	Mu K_i	HLM	MDR	ClogD
26	X_1=N			54	932	30	1.52	2.31
27	X_1=N			72	>3000	39.6	2.32	2.13
28	X_1=CF			10	378	26.2	1.55	3.98
29	X_1=CF			8.8	112	>304	1.50	3.58
30	X_2=N			157	763	20.7	1.45	1.89
31	X_2=N			84	>3000	35.8	1.46	1.83
32	X_3=N			66	>1000	<8	2.01	1.74
33	X_3=N			26	>3000	28.7	1.63	1.85
34	X_4=N			584	>3000	31.4	1.56	1.31
35	X_4=N			75	1327	-	-	2.0
36	X_5=N			>222	>3000	38.3	1.88	1.77
37	X_6=N			54	163	<8	1.86	1.72
38	X_6=N			53	735	<8	1.89	1.85
39	X_6=N			8.4	39	30.9	3.44	2.0

[a] Human κ and μ K_i: Potency reported in nM as average of n = 3. HLM human liver microsomal clearance in (mL/min)/kg. MDR is the efflux ratio BA/AB.

was to synthesize the vast majority of the molecules that only contained 0−1 hydrogen bond donors, had a molecular weight of <425 g/mol, and had a ClogD < 3. By reduction of the number of hydrogen bond donors and lowering of the molecular weight, it would decrease the probability that molecules would be P-gp efflux substrates, thereby allowing penetration into the CNS.[12] Targeting lower lipophilicity than the HTS hit **5** would increase our odds of having improved human liver microsomal clearance and safety.

With the design plan in place the library chemistry was executed by coupling the sulfonyl chlorides with primary and secondary amines. This was followed by a Suzuki coupling utilizing Pd(dppf)Cl_2 as the catalyst in the reaction. Subsequent reductive amination reducing with sodium triacetoxy borohydride provided

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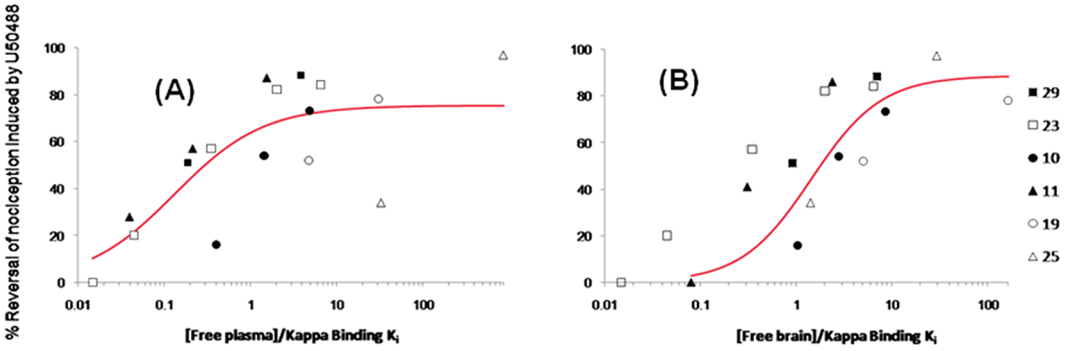

Figure 3. Exposure−effect relationship of compounds **10**, **11**, **19**, **23**, **25**, and **29** dosed subcutaneously: (A) Relationship between free plasma normalized to κ binding K_i and antinociception for κ antagonists; (B) relationship between free brain normalized to κ binding K_i and antinociception for κ antagonists. Symbols represent data from Sprague−Dawley rats. Lines represent the fit of the PK/PD model to the antinociception serum concentrations. Data are presented as mean values [serum concentration (n = 3−4); antinociception (n = 6)].

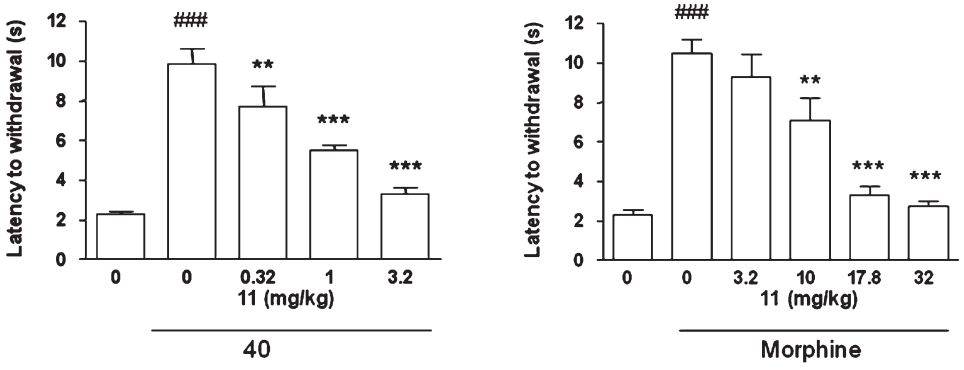

Figure 4. Compound **11** in the tail flick mouse analgesia model response assay vs κ and μ agonists dosed subcutaneously.

the target analogues. This library protocol was successful in producing over 80% of the molecules that were designed.

The library chemistry provided a wealth of SAR knowledge. The biphenyl group provided numerous examples with increased potency and concomitant improvements in physicochemical properties. Molecular weight was reduced significantly in going from the initial R_1 indanol to simple branched alkyls, substituted piperidines, and bicyclic amines. The SAR of the sulfonamide established that 2-substituted piperidines, morpholines, and pyrrolidines along with unsubstituted pyrolidines provided high potency with improved selectivity over μ (Table 1). Compound **8** with a monosubstituted sulfonamide significantly lost κ potency. As designed, a general trend for the whole series was seen that as ClogD was reduced to <2, compounds had low to moderate in vitro human liver microsomal (HLM) clearance. This represented a significant improvement over the initial HTS lead **5**. As was also designed, reducing the hydrogen bond donor count and reducing the molecular weight delivered compounds with reduced P-gp liability. The vast majority of the compounds had a MDR efflux ratio of less than 2. Compound **25** nicely highlights that as two hydrogen bond donors were added, a significant P-gp liability was seen with a MDR efflux ratio of 14. Multiple compounds (**7**, **11**, and **17**) from this initial library set met the project goals of having high potency of <10 nM, low to moderate HLM clearance, and a MDR ratio of <2.5.

The library enablement also allowed for rapid exploration of heterocyclic replacements of the biphenyl (Table 2). The heterocycles were designed to reduce the HLM clearance and potentially maintain the potency and improve the selectivity at μ. If this was obtained, these analogues may have improved safety profiles, lower promiscuity, improved lipophilic ligand efficiency, and improved dose projections. As a general trend the pyridyl analogues had significantly lower HLM clearance than their biphenyl counterparts. For example, compound **37** is not turned over in HLM and compound **11** has a modest clearance of 27 (mL/min)/kg. As expected, the addition of hydrogen bond acceptors did not increase the MDR P-gp efflux liability unlike the addition of hydrogen bond donors.[31] Unfortunately while the pyridyl analogues improved the physicochemical properties and in vitro ADME, a loss in potency and selectivity was seen. When X_{1-5} was nitrogen, a significant loss in κ potency resulted. With X_6 as a nitrogen, compounds (**37**−**39**) had reasonable potency but displayed minimal selectivity (~3×) over μ.

To understand the relationship of in vitro potency to in vivo efficacy, a simplified PK/PD model was sought to understand rodent free brain and free plasma drug levels normalized to the κ K_i. The tail flick analgesia model provided the opportunity to understand in vitro potency to in vivo exposure−effect relationship and to increase confidence in κ pharmacology of this series utilizing **40** (2-(3,4-dichlorophenyl)-*N*-methyl-*N*-[(1R,2R)-2-pyrrolidin-1-ylcyclohexyl]acetamide, U50488H) as a selective κ agonist.[32]

To determine an in vitro potency to in vivo response correlation across compounds from this series, discrete single doses of compounds (Figure 3) were administered to male SD rats followed by a single dose of **40** to the same animals 1 h after dose of

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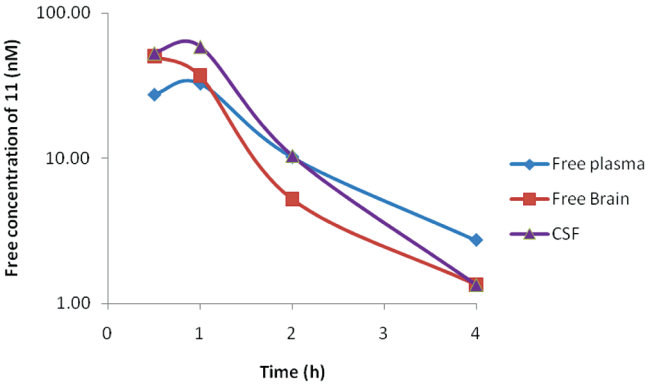

Figure 5. Free brain, free plasma, and CSF drug exposure time course of **11**. Sprague−Dawley rats were dosed at 3.2 mg/kg sc.

administration of the test compounds. Antinociceptive response (hot-plate method) was determined against vehicle groups 30 min after dosing of **40**. Brain tissue and plasma were collected and analyzed for drug exposure using LC−MS/MS. Protein binding of each of the compounds[33] was determined using equilibrium dialysis to calculate free drug exposure in plasma and brain. To understand the concentration−effect relationship for these analogues, reversal of pain tolerance due to κ agonist **40** was plotted using SigmaPlot and fitted to a sigmoidal Emax model against free plasma or free brain exposures normalized to κ binding K_i as shown in Figure 3.

As seen in part A, free plasma exposures vs K_i plot, efficacy did not correlate perfectly with the binding K_i. Compound **25** was the biggest outlier and is not surprising, since it is a P-gp substrate and subject to active efflux due to impediment at the blood−brain barrier. On the other hand, free brain exposures correlated strongly with binding of all compounds considered within this series. This observation is consistent with that made by Kalvass et al., where free brain concentrations (measured in mice dosed with clinically relevant opioids) have shown strong correlation with binding K_i; furthermore, the human EC_{50} for the same set of opioids strongly correlated with the free brain EC_{50} measured in mice (r^2 = 0.95).[34] This understanding of the exposure response relationship demonstrated a functional knowledge that the primary determinant of in vivo effect across this series is indeed κ mediated.

In addition to the exposure response analysis, the tail flick assay has the ability to show functional selectivity of κ vs μ (Figure 4). **11** (2-methyl-*N*-((2′-(pyrrolidin-1-ylsulfonyl)biphenyl-4-yl)-methyl)propan-1-amine, PF-4455242)[35] was tested for its ability to block the analgesic effects of the κ agonist **40** and μ agonist morphine. The ED_{50} to block the κ agonist was 0.67 mg/kg vs 12.0 mg/kg for the μ agonist. This clearly demonstrated the translatability of the in vitro selectivity data to an in vivo model.

From analysis of the PK/PD effects and taking into account the potency, selectivity over μ, physicochemical properties, P-gp liability, human liver microsomal clearance, and binding efficiencies, **11** met our desired criteria for advancement. Compound **11** is a 3 nM antagonist and 1.2 nM in the GTP-γS assay at the κ receptor with over 20-fold selectivity over μ. In addition it possessed the desired in vitro ADME properties with low−moderate human liver microsomal clearance, was not a P-gp substrate with an efflux ratio less than 2.5 in the MDR cell line, and possessed an acceptable HERG therapeutic index. The compound has excellent binding efficiencies with a lipophilic ligand efficiency (LLE) of 6.20 and a ligand efficiency (LE) of 0.33. These properties are all consistent with the marketed CNS drugs.[23]

Table 3. Summary of Preclinical Pharmacokinetic Parameters of Compound 11[a]

parameter	rat	dog	monkey	human
microsomal CL_b[b] ((mL/min)/kg)	44.6	37.7	33	3.1
in vivo CL_b[c] ((mL/min)/kg)	161	29.6	23.8	na
ivivc	3.5×	∼1×	0.79×	na
microsomal ER_h	0.64	0.91	0.68	0.11

[a] na = not applicable. ivivc = in vitro to in vivo correlation. [b] Calculated using the well-stirred model with all binding factors. [c] CL_h determined from JVC rat, dog, and monkey study, iv dose 1 mg/kg.

Further pharmacokinetic profiling showed that **11** has good solubility (>8.53 mg/mL at pH 5.5 and 0.11 mg/mL at pH 6.6.) and permeability in PAMPA and RRCK models (∼19 × 10^{-6} cm/s). The in vitro, in vivo, and in silico data generated preclinically for **11** suggest it will be well absorbed in humans following oral administration. Compound **11** showed good brain penetration in rats (AUC_{0-4h} free brain/free plasma of ∼1, AUC_{0-4h} CSF/unbound plasma of 1.2, AUC_{0-4h} CSF/free brain of ∼1.4) with no evidence of impairment for the compound to cross the blood−brain barrier (Figure 5). It showed moderate in vivo clearance in dog, monkey, and human hepatocytes in vitro; in rats this compound was highly metabolized (Table 3). Overall, **11** was predicted to have moderate human systemic clearance using preclinical methods of clearance predictions.[36] Further details on preclinical and clinical pharmacokinetics and mechanistic studies on phenotyping will be in a future publication.

CONCLUSION

In summary, parallel chemistry enablement and physicochemical property based design were utilized to identify a series of biphenylamine compounds with selectivity for the κ opioid receptor and with druglike properties. The antagonist effects of the molecules were confirmed in testing the ability to block agonist induced analgesia in the tail flick assay and additionally showing functional selectivity for κ over the μ opioid system. An exposure−response analysis was completed in the early stages of the project, clearly demonstrating that central exposure of the compounds in relation to the κ binding affinity was driving efficacy. From this effort, **11** was advanced into broad based pharmacokinetic testing and showed the potential to have desirable pharmacokinetics in humans. This compound has been profiled in multiple models that may be predictive of antidepressive activity, and these data will be disclosed in future publications. With desirable preclinical data, compound **11** has entered into phase 1 human clinical trial and has demonstrated clear human target engagement.[35,37]

EXPERIMENTAL SECTION

All reagents and solvents were used as purchased from commercial sources. Reactions were carried out under a blanket of nitrogen. Silica gel chromatography was done using the appropriate size Biotage prepacked silica filled cartridges. Mass spectral data were collected on a Micromass ADM atmospheric pressure chemical ionization instrument (LRMS APCI). NMR spectra were generated on a Varian 400 MHz instrument. Chemical shifts were recorded in ppm relative to tetramethylsilane (TMS) with multiplicities given as s (singlet), bs (broad singlet), d

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(doublet), t (triplet), dt (doublet of triplets), m (multiplet). Compound purity is determined by combustion analysis (Quantitative Technologies Inc.) or high pressure liquid chromatography (HPLC). HPLC conditions utilized are as follows: gradient, 0−0.25 min 5% A/95% B, 0.25−6.25 min 5% A/95% B → 90% A/10% B, 6.25−6.75 min 90% A/10% B, 6.75−6.85 min 90% A/10% B → 5% A/ 95% B, 6.85−9.5 min 95% A/5% B; column temperature of 45 °C; UV detector, 210 nM. Retention times (t_R) are in minutes, and purity is calculated as % total area. (Column 1: Waters BEH C8 2.1 mm × 100 mm, 1.7 μm. Mobile phase A: acetonitrile. B: 0.1% (v/v) H_3PO_4 + 50 mM $NaClO_4$. Column 2: Waters BEH C8 2.1 mm × 100 mm, 1.7 μm. Mobil phase A: acetonitrile. B: 20 mM ammonium bicarbonate, pH 6.8. Column 3: Waters BEH RP C18 2.1 mm × 100 mm, 1.7 μm. Mobile phase A: acetonitrile. B: 0.1% methanesulfonic acid. Column 4: Waters HSS T3 2.1 mm × 100mm, 1.8 μm. Mobile phase A: acetonitrile. B: 0.1% methanesulfonic acid. Column 5: Waters BEH C8 2.1 mm × 100 mm, 1.7 μm. Mobile phase A: acetonitrile. B: 20 mM ammonium bicarbonate, pH 8.0. All final compounds either met combustion analysis within ±0.4% or were >95% pure by HPLC by the methods above.

General Parallel Procedure for the Preparation of Compounds 6−39. *Step 1. Sulfonamide Formation.* Sulfonyl chloride **A** (0.1 mmol, 1 equiv) and triethylamine (0.2 mmol, 2 equiv) were combined in 0.6 mL of dry methylene chloride, and the amine **B** (0.1 mmol, 1 equiv) in 0.2 mL of dry dichloromethane was added. The mixture was shaken at room temperature for 30 h. Dichloromethane (3 mL) and water (2 mL) were added. The organic layer was washed and transferred to a collection vial. The solvent was removed, and the crude was taken on to next step without further purification.

Step 2. Suzuki Reaction. N_2 was bubbled through a 2 M solution of sodium carbonated and 1,2-dimethoxyethane to deoxygenate. The boronic acids **C** (0.2 mmol, 2 equiv) and the crude bromosulfonamides (0.1 mmol, 1 equiv) were added to 0.6 mL of 1,2-dimethoxyethane. $Pd(dppf)Cl_2$ (0.02 mmol, 0.2 equiv) was added with solid dispenser followed by the addition of 0.6 mL of the 2 M sodium carbonate solution (0.3 mmol, 3 equiv). The mixtures were shaken and heated at 80 °C overnight. Then 2.5 mL of ethyl acetate and 0.5 mL of water were added to the reaction mixture and the organic layer was transferred to a collection vial. The solvent was removed and the crude taken on to next step without purification.

Step 3. Reductive Amination. To the amine **D** (0.12 mmol, 1.2 equiv) was added crude aldehyde (0.1 mmol, 1 equiv) dissolved in 0.5 mL of dry dichloromethane. Sodium triacetoxyborohydride (~0.3 mmol, 3 equiv) was added neat with a solid dispenser. The mixtures were shaken at room temperature overnight. The reaction mixtures were partitioned between 2 mL of 10% ammonium hydroxide and 2.5 mL of dichloromethane. The organic layer was loaded onto a SCX SPE (6 mL, 1 g, SiliCycle) column. The resin was washed with 5 mL of methanol. The compounds were eluted from the resin with 7.5 mL of 10% triethylamine in methanol. The solvent was evaporated and the crude purified via HPLC to provide the library compounds in >95% purity and confirmed by MS.

Singleton Resynthesis for in Vivo Tail Flick Experiments. *1-(2-Bromophenylsulfonyl)pyrrolidine.* Triethylamine (2.0 g, 23.5 mmol) was added to a solution of pyrrolidine (1.67 g, 23.5 mmol) in anhydrous dichloromethane (100 mL, 0.2M). This was followed by dropwise addition of 2-bromobenzenesulfonyl chloride (5.0 g, 20.0 mmol), and the reaction mixture was stirred overnight at room temperature. The reaction mixture was washed with 1 N HCl (2 × 20 mL), and the organic layer was extracted with dichloromethane and dried over $MgSO_4$. The solution was filtered, concentrated, and flash-chromatographed on silica gel, eluting with 30% ethyl acetate/heptane to provide the title compound as an oil (4.0 g, 70%). ^{1}H NMR (400 MHz, $CDCl_3$) δ ppm 8.12 (dd, J = 7.8, 1.8 Hz, 1H), 7.75 (dd, J = 7.8, 1.3 Hz, 1H), 7.36 (m, 2H), 3.37 (m, 4H), 1.87 (m, 4H); GC/MS (M^+ m/z = 290).

2′-(Pyrrolidin-1-ylsulfonyl)biphenyl-4-carbaldehyde. To a solution of 1-(2-bromophenylsulfonyl)pyrrolidine (20 g, 69 mmol) and 4-formylphenylboronic acid (12.90 g, 86.20 mmol) in DME (200 mL) was added 2 M Na_2CO_3 (40 mL) and $Pd(dppf)_2Cl_2$ (563 mg, 0.689 mmol). The mixture was refluxed for 4 h. The reaction mixture was cooled to room temperature and diluted with ethyl acetate (600 mL). It was then washed with 2 M Na_2CO_3 solution (1 × 100 mL), brine (1 × 100 mL), dried over Na_2SO_4, filtered, and concentrated in vacuo. Flash chromatography eluting with 40% ethyl acetate/heptanes provided the title compound (22 g, 69%) as a white solid. ^{1}H NMR (400 MHz, $CDCl_3$) δ ppm 10.06 (s, 1 H), 8.12 (dd, J = 7.9, 1.3 Hz, 1 H), 7.92 (d, J = 7.8 Hz, 2 H), 7.59 (d, J = 7.8 Hz, 2 H), 7.56 (m, 2 H), 7.29 (dd, J = 7.3, 1.3 Hz, 1 H), 2.94 (m, 4 H), 1.40 (m, 4 H); LC/MS (M^+ m/z = 315).

*2-Methyl-N-((2′-(pyrrolidin-1-ylsulfonyl)biphenyl-4-yl)methyl)propan-1-amine (**11**).* 2′-(Pyrrolidin-1-ylsulfonyl)biphenyl-4-carbaldehyde (19.5 g, 61.80 mmol) was dissolved in dichloromethane (200 mL, 0.3 M) followed by the addition of 2-methylpropan-1-amine (5.40 g, 73.80 mmol), and the resulting mixture was stirred for 30 min. Sodium triacetoxyborohydride (19.6 g, 92.30 mmol) was added, and the reaction mixture was stirred overnight. The reaction was quenched with water (50 mL), and the mixture was allowed to stir for 10 min after which the aqueous layer was made basic by adding 10% ammonium hydroxide solution. The solution was extracted with ethyl acetate (3 × 100 mL). The organic layer was dried over sodium sulfate, filtered, and concentrated. The crude product was chromatographed on silica gel using 5−20% methanol/dichloromethane gradient and to provide the title compound (18.28 g, 79%) as an oil. The oil was dissolved in 150 mL of ether, and 15 mL of 2 M HCl−ether solution was added to form a precipitate. The solid was filtered to provide the hydrochloride salt of the title compound as a white solid. ^{1}H NMR (400 MHz, $CDCl_3$) δ ppm 9.92 (S, 1 H), 8.11 (dd, J = 7.8, 1.6 Hz, 1 H), 7.72 (d, J = 7.8 Hz, 2 H), 7.61 (m, 1 H), 7.52 (dd, J = 7.80, 1.6 Hz, 1 H), 7.48 (d, J = 8.2 Hz, 2 H), 7.25 (dd, J = 7.4, 1.6 Hz, 1 H), 4.25 (s, 2 H), 2.90 (m, 4 H), 2.64 (s, 2 H), 2.28 (spt, J = 6.70 Hz, 1 H), 1.77 (m, 4 H), 1.09 (d, J = 6.6 Hz, 6 H); MS (M^+H m/z = 373). Anal. Calcd for $C_{21}H_{29}N_2O_2SCl$: C, 61.67%; H, 7.51%; N, 6.85%. Found C, 61.40%; H, 7.34%; N, 6.75%.

*N-Isopropyl-N-methyl-4′-(piperidin-1-ylmethyl)biphenyl-2-sulfonamide (**10**).* Following the procedure for the preparation of 2-methyl-*N*-((2′-(pyrrolidin-1-ylsulfonyl)biphenyl-4-yl)methyl)propan-1-amine (**11**) but substituting piperidine and 4′-formyl-*N*-isopropyl-*N*-methylbiphenyl-2-sulfonamide (commercial ASDI) provided the title compound (70%). ^{1}H NMR (400 MHz, methanol-d_4) δ ppm 8.07 (d, J = 7.9, 2 H), 7.68 (t, J = 6.2 Hz, 1 H), 7.54 (m, 5 H), 7.34 (d, J = 7.2 Hz, 1 H), 4.34 (s, 2 H), 3.69 (m, 1 H), 3.49 (m, 2 H), 3.03 (dt, J = 12.5, 2.1 Hz, 2 H), 2.31 (s, 3 H), 1.98 (m, 2 H), 1.77 (m, 3 H), 1.53 (m, 1 H), 0.93 (d, J = 6.7, 6 H); MS (M^+H m/z = 386). HPLC purity, >95%.

*N,N-Dimethyl-N-((2′-(pyrrolidin-1-ylsulfonyl)biphenyl-4-yl-methyl)-ethane-1,2-diamine (**19**).* Following the procedure for the preparation of 2-methyl-*N*-((2′-(pyrrolidin-1-ylsulfonyl)biphenyl-4-yl)methyl)propan-1-amine (**11**) but substituting *N*,*N*-dimethylehtane-1,2-diamine provided the title compound (75%). ^{1}H NMR (400 MHz, methanol-d_4) δ ppm 8.03 (dd, J = 7.9, 1.3 1 H), 7.67 (dt, J = 7.5, 1.3 Hz, 1 H), 7.58 (dd, J = 7.9, 1.3 1 H), 7.53 (m, 2 H), 7.44 (m, 2 H), 7.32 (dd, J = 7.9, 1.6 1 H), 4.16 (s, 2 H), 3.21 (m, 2 H), 3.13 (m, 2 H), 2.87 (m, 4 H), 2.70 (s, 6 H), 1.70 (m, 4 H); MS (M^+H m/z = 387). HPLC purity, >95%.

*(±)-6-((2′-(Pyrrolidin-1-ylsulfonyl)biphenyl-4-yl)methyl)-6-azabicyclo-[3.2.1]octane (**23**).* Following the procedure for the preparation of 2-methyl-*N*-((2′-(pyrrolidin-1-ylsulfonyl)biphenyl-4-yl)methyl)propan-1-amine (**11**) but substituting 7-azabicyclo[4.2.1]nonane provided the title compound (83%). ^{1}H NMR (400 MHz, methanol-d_4) δ ppm 8.04 (dd, J = 7.9, 1.3 Hz, 1 H), 7.54 (m, 3 H), 7.50 (d, J = 8.3 Hz, 2 H), 7.34 (dd, J = 7.5, 1.3 Hz, 1 H), 4.52 (m, 2 H), 3.88 (m, 1 H), 3.71 (m, 1 H), 3.36 (m, 1 H), 3.33 (s, 1 H), 2.88 (m, 4 H), 2.70 (s, 1 H), 2.37 (m, 1 H), 2.11 (d, J = 14.1 Hz, 1 H), 1.62 (m, 9 H); MS (M^+H m/z = 411). HPLC purity, >95%.

*(S)-2-(1-((2′-(3-(Hydroxymethyl)piperidin-1-ylsulfonyl)biphenyl-4-yl)methyl)piperidin-4-yl)ethanol (**25**).* Following the procedure for the preparation of 2-methyl-*N*-((2′-(pyrrolidin-1-ylsulfonyl)biphenyl-4-yl)-methyl)propan-1-amine (**11**) but substituting 2-(piperidin-4-yl)ethanol and 2′-(2-(hydroxymethyl)piperidin-1-ylsulfonyl)biphenyl-4-carbaldehyde (commercial ASDI) provided the title compound (83% yield). This compound was separated on a preparative Chiralpack AD column using heptane/ethanol/1% TFA, 80:20, as eluant to afford (*R*)-enatiomer (41% yield, first eluting product, retention time of 21 min) and hydrochloride salts as a white solid. $[\alpha]$ −14.92° (*c* 1, $CHCl_3$). ^{1}H NMR (400 MHz, $CDCl_3$) δ ppm 8.17 (dd, *J* = 7.9, 1.3 Hz, 1 H), 7.52 (m, 1 H), 7.41 (m, 3 H), 7.33 (m, 2 H), 7.30 (d, *J* = 7.5 Hz, 1 H), 3.64 (m, 2 H), 3.61 (d, *J* = 16.6 Hz, 2 H), 3.59 (s, 2H), 3.35 (m, 2 H), 2.88 (m, 2 H), 2.61 (m, 2 H), 2.06 (s, 2 H), 1.69 (d, *J* = 12.9 Hz, 2 H), 1.51 (q, *J* = 6.4 Hz, 4 H), 1.30 (m, 4 H), 1.03 (m, 2 H); MS (M^+H *m*/*z* = 473). HPLC purity, >95%.

1-(2-Bromopheylsulfonyl)piperidine. Following the procedure for the preparation of 1-(2-bromophenylsulfonyl)pyrrolidine but substituting piperidine provided the title compound (89%). ^{1}H NMR (400 MHz, $CDCl_3$) δ ppm 8.07 (dd, *J* = 7.9, 2.1 Hz, 1H), 7.73 (dd, *J* = 7.9, 1.3 Hz, 1H), 7.42 (m, 2H), 3.24 (m, 4H), 1.60 (m, 4H) 1.53 (m, 2H); MS (M^+ *m*/*z* = 305).

3-Fluoro-2′-(piperidin-1-ylsulfonyl)biphenyl-4-carbaldehyde. Following the procedure for the preparation of 2′-(pyrrolidin-1-ylsulfonyl)-biphenyl-4-carbaldehyde but substituting 1-(2-bromophenylsulfonyl)-piperidine and 3-fluoro-4-formylphenylboronic acid provided the title compound (56%). ^{1}H NMR (400 MHz, $CDCl_3$) δ ppm 10.39 (s, 1H), 8.08 (dd, *J* = 7.9, 1.3 Hz, 1H), 7.89 (t, *J* = 7.5 Hz, 1H), 7.62 (dt, *J* = 7.5, 1.7 Hz, 1H), 7.56 (dt, *J* = 7.9, 1.7 Hz, 1H), 7.30 (m, 3H), 2.82 (m, 4H), 1.41 (m, 4H), 1.24 (m, 2H); MS (M^+ *m*/*z* = 348).

*2-(1-((3-Fluoro-2′-(piperidin-1-ylsulfonyl)biphenyl-4-yl)methyl)piperidin-4-yl)ethanol (**29**).* Following the procedure for the preparation of 2-methyl-*N*-((2′-(pyrrolidin-1-ylsulfonyl)biphenyl-4-yl)methyl)propan-1-amine (**11**) but substituting 2-(piperidin-4-yl)ethanol and 3-fluoro-2′-(piperidin-1-ylsulfonyl)biphenyl-4-carbaldehyde provided the title compound (45%). ^{1}H NMR (400 MHz, methanol-d_4) δ ppm 8.07 (dd, *J* = 7.9, 1.3 Hz, 1 H), 7.74 (dt, *J* = 7.5, 1.3 Hz, 1 H), 7.62 (m, 3 H), 7.36 (m, 3 H), 4.42 (s, 2 H), 3.64 (m, 2 H), 3.61 (t, *J* = 6.62 Hz, 2 H), 3.57 (m, 3 H), 3.12 (t, *J* = 12.5 Hz, 2 H), 2.88 (m, 4 H), 2.04 (m, 2 H), 1.77 (m, 1 H), 1.45 (m, 9 H); MS (M^+H *m*/*z* = 460). HPLC purity, >95%.

■ ASSOCIATED CONTENT

Supporting Information. Experimental procedures for the pharmacokinetic experiments, tail flick assay, and binding assays. This material is available free of charge via the Internet at http://pubs.acs.org.

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■ ABBREVIATIONS USED

SAR, structure−activity relationship; VTA, ventral tegmental area; nAcc, nucleus accumbens; BUP, buprenorphine; ADME, absorption, distribution, metabolism, and elimination; LE, ligand efficiency; LLE, lipophilic ligand efficiency; P-gp, P-glycoprotein; MDR, multiple drug resistance; HTS, high throughput screen; HLM, human liver microsomal; PK/PD, pharmacokinetic/pharmacodynamic; PAMPA, parallel artificial membrane permeation assay; AUC, area under the curve; hERG, human ether-a-go-go related gene; RRCK, Russ Ralph canine kidney cells

■ REFERENCES

(1) Dhawan, B. N.; Cesselin, F.; Raghubir, R.; Reisine, T.; Bradley, P. B.; Portoghese, P. S.; Hamon, M. International Union of Pharmacology. XII. Classification of Opioid Receptors. *Pharmacol. Rev.* **1996**, *48*, 567–592.

(2) Aldrich, J. V.; Vigil-Cruz, S. C. Narcotic Analgesics. In *Burger's Medicinal Chemistry and Drug Discovery*; Abraham, D., Ed.; Wiley: Hoboken, NJ, 2003.

(3) Coop, A.; Rice, K. C. Role of Delta-Opioid Receptors in Biological Processes. *Drug News Perspect.* **2000**, *13*, 481–487.

(4) Pfeiffer, A.; Brantl, V.; Herz, A.; Emrich, H. M. Psychotomimesis Mediated by Kappa Opiate Receptors. *Science* **1986**, *233*, 774–776.

(5) Newton, S. S.; Thome, J.; Wallace, T. L.; Shirayama, Y.; Schlesinger, L.; Sakai, N.; Chen, J.; Neve, R.; Nestler, E. J.; Duman, R. S. Inhibition of cAMP Response Element-Binding Protein or Dynorphin in the Nucleus Accumbens Produces an Antidepressant-like Effect. *J. Neurosci.* **2002**, *22*, 10883–10890.

(6) Emrich, H. M.; Vogt, P.; Herz, A. Possible Antidepressive Effects of Opioids: Action of Buprenorphine. *Ann. N.Y. Acad. Sci.* **1982**, *398*, 108–112.

(7) Kosten, T. R.; Morgan, C.; Kosten, T. A. Depressive Symptoms during Buprenorphine Treatment of Opioid Abusers. *J. Subst. Abuse Treat.* **1990**, *7*, 51–54.

(8) Mague, S. D.; Pliakas, A. M.; Todtenkopf, M. S.; Tomasiewicz, H. C.; Zhang, Y.; Stevens, W. C.; Jones, R. M.; Portoghese, P. S; Carlezon, W. A. Antidepressant-like Effects of κ-Opioid Receptor Antagonists in the Forced Swim Test in Rats. *J. Pharmacol. Exp. Ther.* **2003**, *305*, 323–330.

(9) McLaughlin, J. P.; Li, S.; Valdez, J.; Chavkin, T. A.; Chavkin, C. Social Defeat Stress-Induced Behavioral Responses Are Mediated by the Endogenous Kappa Opioid System. *Neuropsychopharmacology* **2006**, *31*, 1241–1248.

(10) Carey, A. N.; Borozny, K.; Aldrich, J. V.; McLaughlin, J. P. Reinstatement of Cocaine Place-Conditioning Prevented by the Peptide Kappa-Opioid Receptor Antagonist Arodyn. *Eur. J. Pharmacol.* **2007**, *569*, 84–89.

(11) Portoghese, P. X.; Lipkowski, A. W.; Takemori, A. E. Binaltorphimin and Nor-binaltorphimin, Potent and Selective Kappa-Opioid Receptor Antagonists. *Life Sci.* **1987**, *40* (13), 1287–1292.

(12) Jones, R. M.; Portoghese, P. S. 5-Guanidinonaltrindole, a Highly Selective and Potent Kappa-Opioid Receptor Antagonist. *Eur. J. Pharmacol.* **2000**, *396* (1), 49–52.

(13) Mitch, C. H.; Zimmerman, D. M.; Snoddy, J. D.; Reel, J. K.; Cantrell, B. E. Synthesis and Absolute Configuration of LY255582, a Potent Opioid Antagonist. *J. Org. Chem.* **1991**, *56* (4), 1660–1663.

(14) Zimmerman, D. M.; Leander, J. D.; Cantrell, B. E.; Reel, J. K.; Snoddy, J.; Mendelsohn, L. G.; Johnson, B. G.; Mitch, C. H. Structure −Activity Relationships of *trans*-3,4-Dimethyl-4-(3-hydroxyphenyl)piperidine Antagonists for μ- and κ-Opioid Receptors. *J. Med. Chem.* **1993**, *36* (20), 2833–2841.

(15) Thomas, J. B.; Fall, M. J.; Cooper, J. B.; Rothman, R. B.; Mascarella, S. W.; Xu, H.; Partilla, J. S.; Dersch, C. M.; McCullough, K. B.; Cantrell, B. E.; Zimmerman, D. M.; Carroll, F. I. Identification of an Opioid κ-Receptor Subtype-Selective N-Substituent for (+)-(3*R*,4*R*)-Dimethyl-4-(3-hydroxyphenyl)piperidine. *J. Med. Chem.* **1998**, *41* (26), 5188–5197.

(16) Thomas, J. B.; Atkinson, R. N.; Rothman, R. B.; Fix, S. E.; Mascarella, S. W.; Vinson, N. A.; Xu, H.; Dersch, C. M.; Lu, Y. F.; Cantrell, B. E.; Zimmerman, D. M.; Carroll, F. I. Identification of the First *trans*-(3*R*,4*R*)-Dimethyl-4-(3-hydroxyphenyl)piperidine Derivative to Possess Highly Potent and Selective Opioid κ Receptor Antagonist Activity. *J. Med. Chem.* **2001**, *44* (17), 2687–2690.

(17) Wager, T. T.; Chandrasekaran, R. Y.; Hou, X.; Troutman, M. D.; Verhoest, P. R.; Villalobos, A.; Will, Y. Defining Desirable Central Nervous System Drug Space through the Alignment of Molecular Properties, in Vitro ADME, and Safety Attributes. *ACS Chem. Neurosci.* **2010**, *1* (6), 420–434.

(18) Hopkins, A. L.; Groom, C. R.; Alex, A. Ligand Efficiency: A Useful Metric for Lead Selection. *Drug Discovery Today* **2004**, *9* (10), 430–431.

(19) Kuntz, I. D.; Chen, K.; Sharp, K. A.; Kollman, P. A. The Maximal Affinity of Ligands. *Proc. Natl. Acad. Sci. U.S.A.* **1999**, *96* (18), 9997–10002.

(20) Thomas, J. B.; Atkinson, R. N.; Vinson, N. A.; Catanzaro, J. L.; Perretta, C. L.; Fix, S. E.; Mascarella, S. W.; Rothman, R. B.; Xu, H.; Dersch, C. M.; Cantrell, B. E.; Zimmerman, D. M.; Carroll, F. I. Identification of (3*R*)-7-Hydroxy-*N*-((1*S*)-1-{[(3*R*,4*R*)-4-(3-hydroxyphenyl)-3,4-dimethyl-1-piperidinyl]methyl}-2-methylpropyl)-1,2,3,4-tetrahydro-3-isoquinolinecarboxamide as a Novel Potent and Selective Opioid κ Receptor Antagonist. *J. Med. Chem.* **2003**, *46* (14), 3127–3137.

(21) Lewis, D. F. V.; Eddershaw, P. J.; Goldfarb, P. S.; Tarbit, M. H. Molecular Modeling of Cytochrome P4502D6 (CYP2D6) Based on an Alignment with CYP102: Structural Studies on Specific CYP2D6 Substrate Metabolism. *Xenobiotica* **1997**, *27* (4), 319–339.

(22) De Groot, M. J.; Ackland, M. J.; Horne, V. A.; Alex, A. A.; Jones, B. C. Novel Approach to Predicting P450-Mediated Drug Metabolism: Development of a Combined Protein and Pharmacophore Model for CYP2D6. *J. Med. Chem.* **1999**, *42* (9), 1515–1524.

(23) Liras, S.; McHardy, S. F.; Allen, M. P.; Segelstein, B. E.; Heck, S. D.; Bryce, D. K.; Schmidt, A. W.; Vanase-Frawley, M.; Callegari, E.; McLean, S. Biaryl Piperidines as Potent and Selective Delta Opioid Receptor Ligands. *Bioorg. Med. Chem. Lett.* **2010**, *20* (2), 503–507.

(24) Wager, T. T.; Chandrasekaran, R. Y.; Hou, X.; Troutman, M. D.; Verhoest, P. R.; Villalobos, A.; Will, Y. Defining Desirable Central Nervous System Drug Space through the Alignment of Molecular Properties, in Vitro ADME, and Safety Attributes. *ACS Chem. Neurosci.* **2010**, *1* (6), 420–434.

(25) Whalen, K.; Gobey, J.; Janiszewski, J. A. Centralized Approach to Tandem Mass Spectrometry Method Development for High-Throughput ADME Screening. *Rapid Commun. Mass Spectrom.* **2006**, *20*, 1497–1503.

(26) Feng, B; Mills, J. B.; Davidson, R. E.; Mireles, R. J.; Janiszewski, J. S.; Troutman, M. D.; de Morais, S. M. In Vitro P-glycoprotein Assays to Predict the in Vivo Interactions of P-Glycoprotein with Drugs in the Central Nervous System. *Drug Metab. Dispos.* **2008**, *36* (2), 268–275.

(27) Hughes, J. D.; Blagg, J.; Price, D. A.; Bailey, S.; DeCrescenzo, G. A.; Devraj, R. V.; Ellsworth, E.; Fobian, Y. M.; Gibbs, M. E.; Gilles, R. W.; Greene, N.; Huang, E.; Krieger-Burke, T.; Loesel, J.; Wager, T.; Whiteley, L.; Zhang, Y. Physiochemical Drug Properties Associated with in Vivo Toxicological Outcomes. *Bioorg. Med. Chem. Lett.* **2008**, *18*, 4872–4875.

(28) Rang, H. P. The Receptor Concept: Pharmacology's Big Idea. *Br. J. Pharmacol.* **2006**, *147*, S9–S16.

(29) Verhoest, P. R.; Proulx-Lafrance, C.; Corman, M.; Chenard, L.; Helal, C. J.; Hou, X.; Kleiman, R.; Liu, S.; Marr, E.; Menniti, F. S.; Schmidt, C. J.; Vanase-Frawley, M.; Schmidt, A. W.; Williams, R. D.; Nelson, F. R.; Fonseca, K. R.; Liras, S. Identification of a Brain Penetrant PDE9A Inhibitor Utilizing Prospective Design and Chemical Enablement as a Rapid Lead Optimization Strategy. *J. Med. Chem.* **2009**, *52* (24), 7946–7949.

(30) Hughes, J. D.; Blagg, J.; Price, D. A.; Bailey, S.; DeCrescenzo, G. A.; Devraj, R. V.; Ellsworth, E.; Fobian, Y. M.; Gibbs, M. E.; Gilles, R. W.; Greene, N.; Huang, E.; Krieger-Burke, T.; Loesel, J.; Wager, T.; Whiteley, L.; Zhang, Y. Physiochemical Drug Properties Associated with in Vivo Toxicological Outcomes. *Bioorg. Med. Chem. Let.* **2008**, *18* (17), 4872–4875.

(31) Wager, T. T.; Hou, X.; Verhoest, P. R.; Villalobos, A. Moving beyond Rules: The Development of a Central Nervous System Multiparameter Optimization (CNS MPO) Approach To Enable Alignment of Druglike Properties. *ACS Chem. Neurosci.* **2010**, *1* (6), 435–449.

(32) Dosaka-Akita, K.; Tortella, F. C.; Holaday, J. W.; Long, J. B. The Kappa Opioid Agonist U-50488H Antagonizes Respiratory Effects of Mu Opioid Receptor Agonists in Conscious Rats. *J. Pharmacol. Exp. Ther.* **1993**, *264* (2), 631–637.

(33) Kalvass, J. C.; Mauer, T. S. Influence of Nonspecific Brain and Plasma Binding on CNS Exposure: Implications for Rational Drug Discovery. *Biopharm. Drug Dispos.* **2002**, *23* (8), 327–338.

(34) Kalvass, J. C.; Olson, E. R.; Cassidy, M. P.; Selley, D. E.; Pollack, G. M. Pharmacokinetics and Pharmacodynamics of Seven Opioids in P-Glycoprotein-Competent Mice: Assessment of Unbound Brain EC50, u and Correlation of in Vitro, Preclinical, and Clinical Data. *J. Pharmacol. Exp. Ther.* **2007**, *323* (1), 346–355.

(35) Jacobsen, L. K.; Banerjee, A.; Byon, W.; Huang, Y.; Carson, R. E.; Tomasi, G.; Weinzimmer, D.; Williams, W.; Lin, S.-F.; Ropchan, J.; McCarthy, T. J.; Grimwood, S.; Badura, L. Central Receptor Occupancy and Pharmacodynamic Action of PF-04455242, a High Affinity, Selective Kappa Opioid Receptor Antagonist, in Humans. *Society for Neuroscience*; Society for Neuroscience: Washington, DC, 2010.

(36) Obach, S. R.; Baxter, J. G.; Liston, T. E.; Silber, B. M.; Jones, B. C.; MacIntyre, F.; Rance, D. J.; Wastall, P. The Prediction of Human Pharmacokinetic Parameters from Preclinical and in Vitro Metabolism Data. *J. Pharmacol. Exp. Ther.* **1997**, *283* (1), 46–58.

(37) Jacobsen, L. K.; Banerjee, A.; Byon, W.; Huang, H.; Carson, R. E.; Weinzimmer, D.; McCarthy, T. J.; Grimwood, S.; Badura, L. Central Receptor Occupancy and Pharmacodynamic Action of PF-04455242, a High Affinity, Selective Kappa Opioid Receptor Antagonist, in Humans. *Society for Neuroscience*; Society for Neuroscience: Washington, DC, 2010.