ACS Chemical Neuroscience

Research Article

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Synthesis, Pharmacological Characterization, and Structure–Activity Relationship Studies of Small Molecular Agonists for the Orphan GPR88 Receptor

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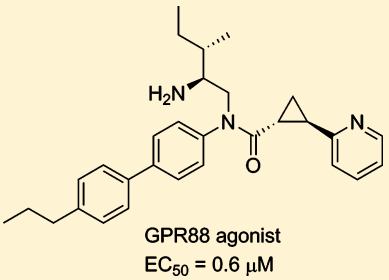
Supporting Information

ABSTRACT: GPR88 is an orphan G-protein-coupled receptor (GPCR) enriched in the striatum. Genetic deletion and gene expression studies have suggested that GPR88 plays an important role in the regulation of striatal functions and is implicated in psychiatric disorders. The signal transduction pathway and receptor functions of GPR88, however, are still largely unknown due to the lack of endogenous and synthetic ligands. In this paper, we report the synthesis of a GPR88 agonist 2-PCCA and its pure diastereomers, which were functionally characterized in both transiently and stably expressing GPR88 HEK293 cells. 2-PCCA inhibited isoproterenol-stimulated cAMP accumulation in a concentration-dependent manner in cells expressing GPR88 but not in the control cells, suggesting that the observed cAMP inhibition is mediated through GPR88 and that GPR88 is coupled to $G\alpha_i$. 2-PCCA did not induce calcium mobilization in GPR88 cells, indicating no $G\alpha_q$-mediated response. A structure–activity relationship (SAR) study of 2-PCCA was also conducted to explore the key structural features for GPR88 agonist activity.



KEYWORDS: *Orphan GPR88, agonists, 2-PCCA*

GPR88 is an orphan G-protein-coupled receptor, which was originally identified as a striatum-specific receptor (designated Strg/GPR88),[1] though it is also expressed in other brain regions, including the cerebral cortex, amygdala, and hypothalamus.[2] In the striatum, GPR88 is highly expressed in both D_1 and D_2 receptor-expressing medium spiny neurons (MSNs),[2b,3] suggesting the receptor may play a role in regulating dopaminergic activity. GPR88 knockout mice demonstrated disrupted prepulse inhibition of the startle response, a phenotype of schizophrenia, and exhibited D_2 receptors hypersensitivity (as evidenced by increased sensitivity to apomorphine-induced climbing and stereotypy, and amphetamine-stimulated locomotor activity).[4] In another study of GPR88 knockout mice,[3,5] the animals exhibited increased locomotion, and impaired motor coordination and cue-based learning. GPR88 re-expression normalized these impaired behaviors, suggesting that GPR88 dysfunction may contribute to abnormal behaviors observed in neurological and psychiatric diseases.[3] In line with these findings from GPR88 knockout studies, transcriptional profiling studies have revealed GPR88 gene expression is altered by treatment or conditions related to schizophrenia,[6] bipolar disorder,[7] depression,[8] and drug addiction.[9] Taken together, these studies suggest that GPR88 plays an important role in the regulation of striatal functions and is a promising drug target for treating basal ganglia-associated disorders.

In order to elucidate the biological function of GPR88, selective agonists are required. Recently, a series of surrogate agonists of GPR88 have been reported in the patent literature and were suggested to activate GPR88 coupling to $G\alpha_i$ pathways.[10] However, the function and structure–activity relationship (SAR) relative to these compounds are unclear. In this paper, we report the synthesis of a GPR88 ligand 2-PCCA [(1*R**,2*R**)-2-(pyridin-2-yl)cyclopropanecarboxylic acid ((2*S*,3*S*)-2-amino-3-methylpentyl)-(4′-propylbiphenyl-4-yl)-amide (**1**); Figure 1] and its pure (1*R*,2*R*)- and (1*S*,2*S*)-diastereomers (**2** and **3**, respectively), which were functionally characterized in the GPR88 cell-based cAMP assays. A series of 2-PCCA analogues (**4a–i**, **5a–e**, **6a**, and **6b**; Figure 2) were also synthesized and examined to explore the SAR of this chemical scaffold at GPR88.

Received: April 14, 2014
Revised: May 2, 2014
Published: May 2, 2014

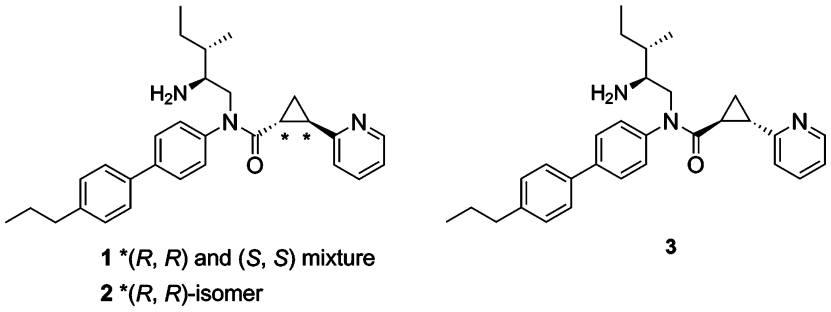


Figure 1. Structures of 2-PCCA (**1**), **2**, and **3**.

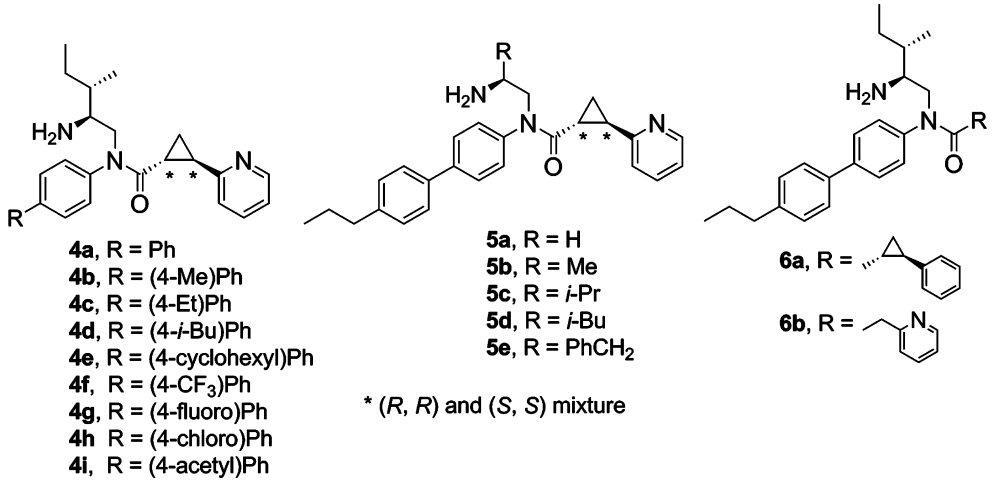


Figure 2. 2-PCCA analogues.

CHEMISTRY

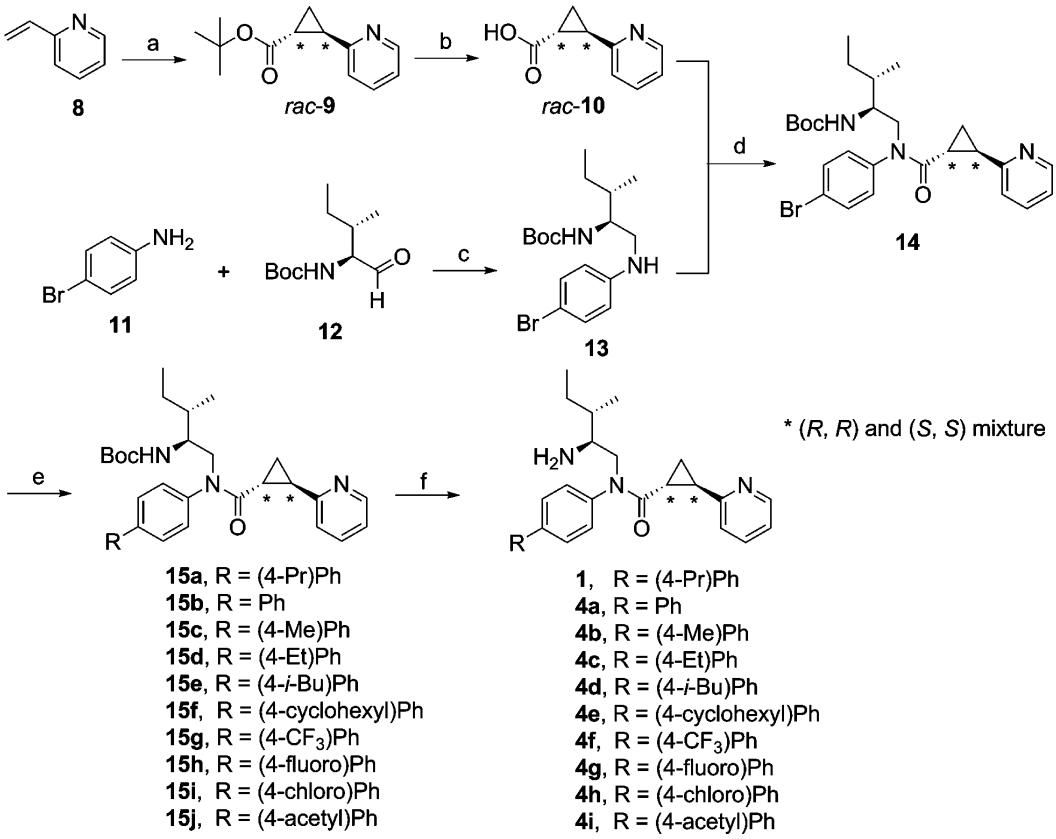
2-PCCA (**1**) and the 4-substituted bisphenyl analogues **4a**−**i** were synthesized by a known procedure,[10b] outlined in Scheme 1, with some modifications. A diastereomeric mixture was first synthesized to characterize the GPR88 signaling pathways. Asymmetric synthesis of both pure diastereomers **2** and **3** of 2-PCCA was conducted later to determine which isomer is more active. Cyclopropanation of 2-vinylpyridine (**8**) with *tert*-butyl diazoacetate under catalytic conditions led to *tert*-butyl ester **9**, which was then treated with 4 M HCl in dioxane to give the racemic (1*R**,2*R**)-2-(pyridin-2-yl)cyclopropanecarboxylic acid (**10**) in 56% yield. Reductive amination of aldehyde **12**, prepared by Dess-Martin oxidation of commercially available (−)-(2*S*,3*S*)-*N*-Boc-2-amino-3-methyl-1-pentanol with 4-bromoaniline (**11**) afforded amine **13** in 75% yield. With both building blocks available, amide **14** was synthesized in 72% yield by converting acid **10** into the corresponding acid chloride, followed by reaction with amine **13**. Suzuki coupling of **14** with an appropriate arylboronic acid under microwave conditions gave intermediates **15a**−**j** in the range of 65−96% yields. Removal of the Boc protecting group with 4 M HCl in dioxane provided **1** and **4a**−**i** in 90−98% yields. Compounds **1** and **4a**−**i** were determined to be 1:1 diastereomeric mixtures by ^{1}H NMR and HPLC analyses.

Synthesis of pure diastereomers **2** and **3** of 2-PCCA is described in Scheme 2. The key intermediate **14**, prepared from enantiomerically pure (−)-(2*S*,3*S*)-*N*-Boc-2-amino-3-methyl-1-pentanol as described in Scheme 1, is a 1:1 mixture of (1*R*,2*R*)- and (1*S*,2*S*)-diastereomers differentiating at the configuration of the *trans*-substituted cyclopropane ring. Asymmetric synthesis of (1*R*,2*R*)-**14** was accomplished starting from the preparation of pure enantiomer (1*R*,2*R*)-2-(pyridin-2-yl)-cyclopropanecarboxylic acid ((1*R*,2*R*)-**10**). Thus, asymmetric cyclopropanation of **8** using the known chiral porphyrin catalyst [Co(3,5-Di^{t}Bu-ChenPhyrin)][11] afforded the *tert*-butyl ester (1*R*,2*R*)-**9** in 97% ee, as determined by chiral HPLC analysis. The chiral porphyrin Co(II) catalysts have been well studied in the asymmetric cyclopropanation of olefins using diazoacetates to give the corresponding cyclopropanes with high diastereoselectivity and enantioselectivity.[11] Assignment of the absolute configuration of (1*R*,2*R*)-**9** was made based on an analogy to the known (1*R*,2*R*)-2-phenyl-1-cyclopropanecarboxylic acid *tert*-butyl ester[11a] synthesized using the same chiral porphyrin catalyst. Acidic hydrolysis of (1*R*,2*R*)-**9** led to acid (1*R*,2*R*)-**10**, which was then coupled with amine **13** to provide (1*R*,2*R*)-**14** in 40% yield over three steps. To obtain the pure diastereomer (1*S*,2*S*)-**14**, the mixture **14** was separated by HPLC using a ChiralPak IA column to afford (1*R*,2*R*)-**14** and (1*S*,2*S*)-**14** in 40% and 39% yield, respectively. Suzuki coupling of (1*R*,2*R*)-**14** and (1*S*,2*S*)-**14** with 4-propylphenylboronic acid, followed by removal of the Boc protecting group with HCl gave **2** and **3**, in 80% and 81% yield, respectively.

Compounds **5a**−**e** were synthesized using the procedure, outlined in Scheme 3, analogous to that used to prepare **1**. Reductive amination of an appropriate aldehyde **17a**−**e** with 4-bromoaniline (**11**) or 4-(4′-propylphenyl)aniline (**16**) afforded amine **18a**−**e** in 50−86% yields. Amide formation with the acid chloride, prepared from the racemic **10**, gave **19a**−**e** in 53−60% yields. Suzuki coupling of **19a**, **19b**, and **19e** with 4-propylphenylboronic acid yielded 53−80% of **20a**, **20b**, and

dx.doi.org/10.1021/cn500082p | ACS Chem. Neurosci. 2014, 5, 576−587

Scheme 1[a]



[a]Reagents: (a) *tert*-butyl diazoacetate, 5,10,15,20-tetraphenyl-21*H*,23*H*-porphine cobalt(II), toluene, 80 °C, 2 h; (b) 4 M HCl/dioxane, DCM, rt, overnight; (c) $NaBH(OAc)_3$, 1,2-dichloroethane, rt, overnight; (d) **10**/oxalyl chloride/DCM/40 °C/2 h, concentrated, then **13**/Et_3N/DCM, rt, overnight; (e) arylboronic acid, Pd(dppf)Cl_2·DCM, K_3PO_4, DME/H_2O (3:1), microwave, 160 °C, 6 min; (f) 4 M HCl/dioxane, DCM, rt, 6 h.

20e. Deprotection of the Boc group furnished **5a**−**e** as 1:1 diastereomeric mixtures in 92−98% yields.

Synthesis of compounds **6a** and **6b** started with the reaction of amine **13** with (1*R*,2*R*)-2-phenyl-1-cyclopropanecarboxylic acid[12] and 2-pyridylacetic acid (Scheme 4), respectively. The resulting amides **21a** and **21b** were coupled with 4-propylphenylboronic acid, followed by HCl treatment to provide **6a** and **6b** in 55% and 33% overall yield, respectively. All synthesized compounds were >95% pure as determined by HPLC analyses. The ^{1}H NMR spectra of the target compounds were in agreement with the assigned structures.

RESULTS AND DISCUSSION

Despite emerging pharmacological implications of the orphan GPR88 receptor, little is known regarding its downstream signaling pathways, as identification of endogenous and synthetic ligands has been elusive. Results from the patent literature[10] indicated that GPR88 couples to $G\alpha_i$ proteins and thereby inhibits cAMP production. To develop an appropriate cell based assay system to support drug discovery for the GPR88 receptor, we initially transiently cotransfected HEK 293T cells with the human GPR88 cDNA and a luminescent cAMP biosensor and determined both $G\alpha_s$ and $G\alpha_i$ activations.[13] 2-PCCA (**1**) produced no measurable increases in cAMP levels at concentrations up to 30 μM. As a positive control, isoproterenol (ISO) activated the endogenous β_2 adrenergic receptors expressed in HEK293T cells and greatly stimulated cAMP production in a concentration-dependent manner. 2-PCCA inhibited ISO-induced cAMP formation with a pEC_{50} value of 6.06 (EC_{50} = 877 nM) in GPR88 cells but not in the control cells transiently transfected with the biosensor, demonstrating that 2-PCCA activates GPR88-mediated $G\alpha_i$ signaling (Figure 3). In addition, the (1*R*,2*R*)-isomer **2** (EC_{50} = 373 nM) is approximately 5-fold more potent than the (1*S*,2*S*)-isomer **3**. Furthermore, 2-PCCA did not induce calcium mobilization measured by the fluorescent imaging plate reader (FLIPR) calcium assay[13b,14] in HEK293T/GPR88 cells (data not shown), indicating that GPR88 is likely not coupled to $G\alpha_q$ proteins in our assay systems.

To further characterize the GPR88 in vitro functions and explore the key structural features of 2-PCCA agonist activity, HEK293 cells stably expressing the human GPR88 receptor and the GloSensor-22F cAMP construct were established. In the stable GPR88-22F cells, 2-PCCA and its pure diastereomer **2** had pEC_{50} values of 6.04 (EC_{50} = 911 nM) and 6.22 (EC_{50} = 603 nM), respectively. To explore the SAR of 2-PCCA, substitution effects of the bisphenyl moiety were first examined. As seen in Table 1, unsubstituted analogue **4a** was less potent than 2-PCCA. The 4-position tolerated small to medium size of alkyl substitutions with the methyl analogue **4b** (EC_{50} = 845 nM) and cyclohexyl analogue **4e** (EC_{50} = 746 nM) being the most potent compounds in the series. Replacing the methyl group with an electron-withdrawing trifluoromethyl group (**4f**) markedly reduced activity. The addition of a fluoro or chloro group to the 4-position led to **4g** and **4h**, respectively, resulting in even lower potency. Somewhat surprisingly, the 4-acetyl

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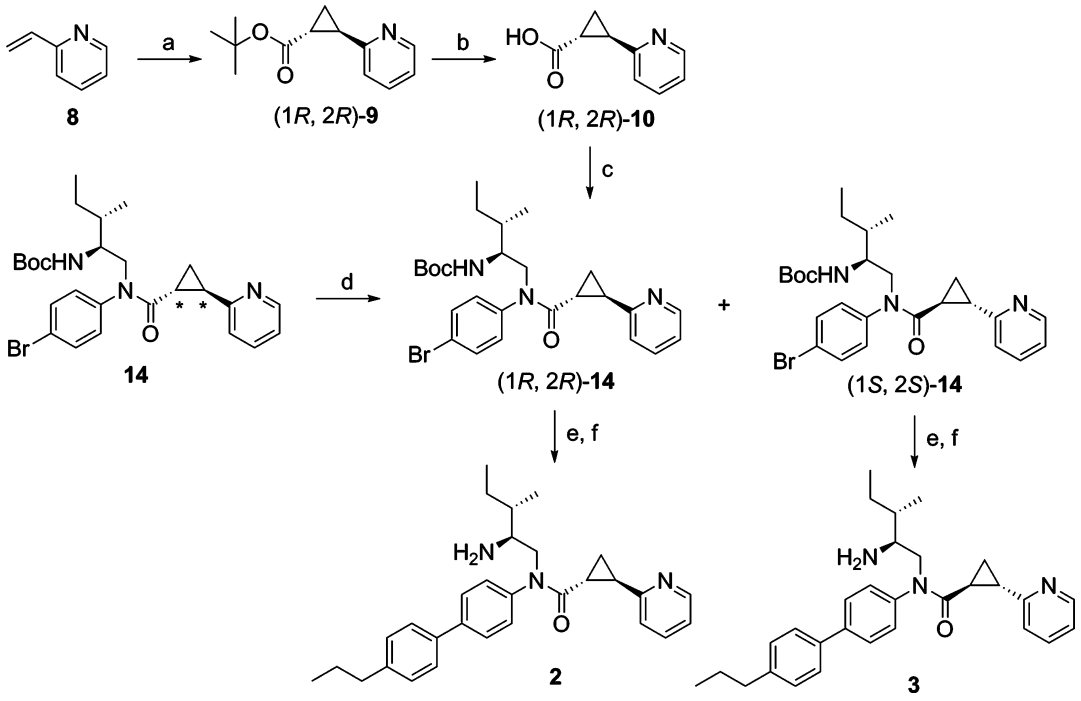
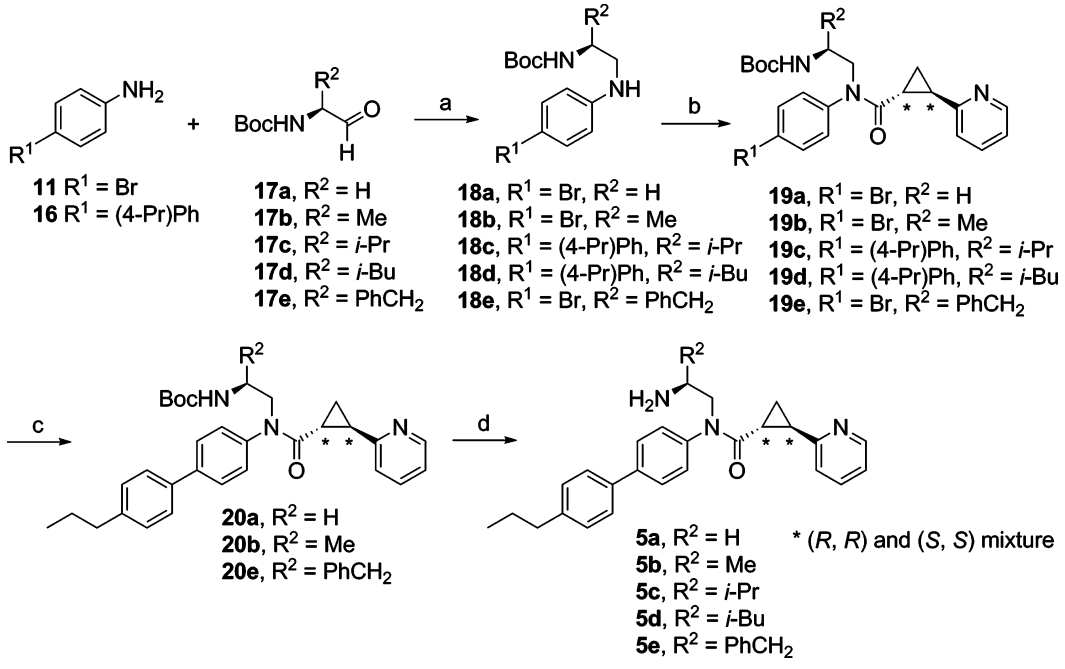
Scheme 2[a]



[a]Reagents: (a) *tert*-butyl diazoacetate, 1 mol % Co(3,5-di-*t*-Bu-ChenPhyrin) catalyst, DMAP, toluene, rt, 48 h; (b) 4 M HCl/dioxane, DCM, rt, overnight; (c) (1*R*,2*R*)-**10**/oxalyl chloride/DCM/40 °C/2 h, concentrated, then **13**/ Et_3N/DCM, rt, overnight; (d) HPLC separation; (e) 4-propylphenylboronic acid, Pd(dppf)Cl_2·DCM, K_3PO_4, DME/H_2O (3:1), microwave, 160 °C, 6 min; (f) 4 M HCl/dioxane, DCM, rt, 6 h.

Scheme 3[a]



[a]Reagents: (a) $NaBH(OAc)_3$, 1,2-dichloroethane, rt, overnight; (b) **10**/oxalyl chloride/DCM/40 °C/2 h, concentrated, then **18a**–**e**/Et_3N/DCM, rt, overnight; (c) 4-propylphenylboronic acid, Pd(dppf)Cl_2·DCM, K_3PO_4, DME/H_2O (3:1), microwave, 160 °C, 6 min; (d) 4 M HCl/dioxane, DCM, rt, 6 h.

analogue **4i** possessed a similar potency (EC_{50} = 923 nM) at GPR88 relative to 2-PCCA.

Investigation of the substituted ethylamine moiety of 2-PCCA (Table 2) showed that hydrophobic substitutions were well tolerated. The trend of increased potency with large substituents (**5a**–**e**) at the ethylamine moiety suggests that a hydrophobic pocket may be present in the GPR88 receptor. A limited examination of the amide carbonyl groups observed that the pyridyl group in 2-PCCA could be replaced with a phenyl group (**6a**) while causing only a slight decrease in potency.

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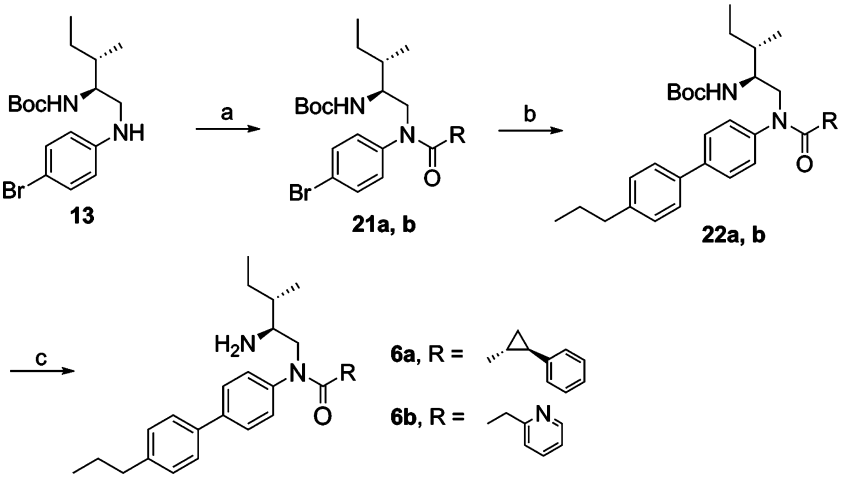
Scheme 4[a]



[a]Reagents: (a) **21a**: (1*R*, 2*R*)-2-phenyl-1-cyclopropanecarboxylic acid/$SOCl_2$/reflux, overnight, concentrated, then **13**/Et_3N/DCM, rt, overnight; **21b**: 2-pyridylacetic acid, HBTU, Et_3N, MeCN, rt, overnight; (b) 4-propylphenylboronic acid, Pd(dppf)Cl_2·DCM, K_3PO_4, DME/H_2O (3:1), microwave, 160 °C, 6 min; (c) 4 M HCl/dioxane, DCM, rt, 6 h.

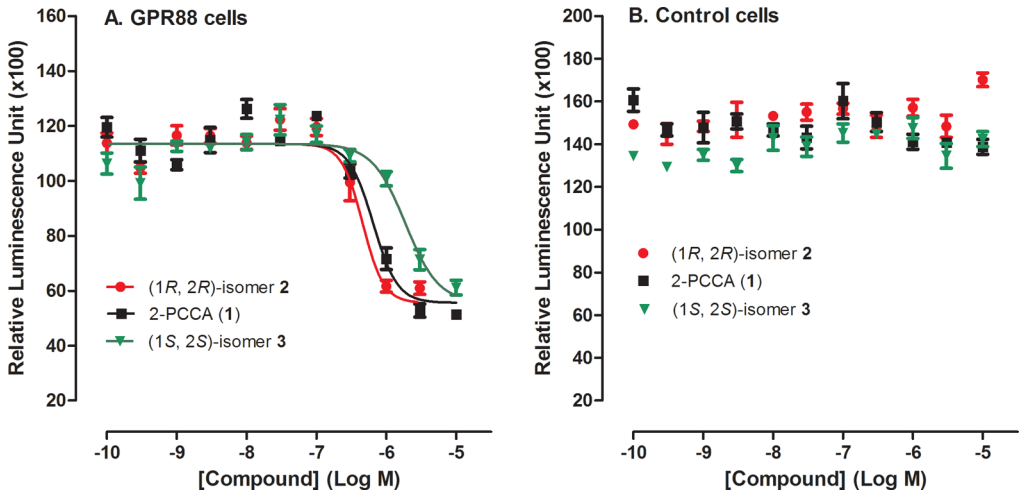


Figure 3. HEK 293T cells were transiently transfected with GPR88 and GloSensor cAMP construct (A) or GloSensor cAMP only (B). 2-PCCA (**1**), **2**, and **3** inhibited isoproterenol-induced cAMP production in GPR88 cells, but not in the control cells. The data are the means of quadruplicate measurements with standard deviation shown as error bars and are representative of at least three independent experiments.

However, replacement of the cyclopropane moiety with a methylene group (**6b**) resulted in a loss of activity, indicating the central linker of the aromatic and carbonyl moieties is critical for GPR88 recognition.

CONCLUSIONS

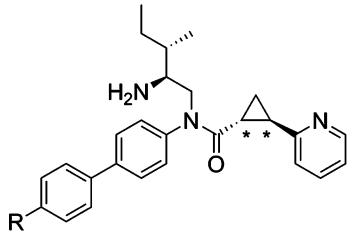
In summary, we demonstrated that GPR88 couples to the $G\alpha_i$ subunits, and is activated by 2-PCCA in both transient and stable GPR88 expressing cells. In an effort to determine the key structural features for 2-PCCA agonist activity, we designed and synthesized a series of 2-PCCA analogues **4a**−**i**, **5a**−**e**, **6a**, and **6b**. Further pharmacological evaluation of the (1*R*,2*R*)-isomer **2** and phenyl analogue **6a**, including receptor specificity and in vivo behavioral studies, are in progress. These studies will facilitate the identification of highly potent, selective ligands for GPR88 and the understanding of its physiological functions in vivo.

METHODS

Chemistry. *General Methods.* Melting points were determined using a MEL-TEMP II capillary melting point apparatus and are uncorrected. Nuclear magnetic resonance (^1H NMR and ^{13}C NMR) spectra were obtained on a Bruker Avance DPX-300 MHz NMR spectrometer. Chemical shifts are reported in parts per million (ppm) with reference to internal solvent. ^{13}C NMR data of diastereomeric mixtures were not reported due to the complicity of the spectra. Mass spectra (MS) were run on a PerkinElmer Sciex API 150 EX mass spectrometer. HRMS spectra were run on a Waters Synapt G2 HDMS Q-TOF mass spectrometer, using electrospray ionization in positive ion mode. Optical rotations were measured on an AutoPol III polarimeter, purchased from Rudolf Research. Analytical thin-layer chromatography (TLC) was carried out using EMD silica gel 60 F_{254} TLC plates. TLC visualization was achieved with a UV lamp or in an iodine chamber. Flash column chromatography was done on a CombiFlash Companion system using Isco prepacked silica gel columns. Unless otherwise stated, reagent-grade chemicals were obtained from commercial sources and were used without further purification. All moisture- and air-sensitive reactions and reagent transfers were carried out under dry nitrogen.

dx.doi.org/10.1021/cn500082p | ACS Chem. Neurosci. 2014, 5, 576−587

Table 1. Structures and Activities of Compounds 1, 2, and 4a−i



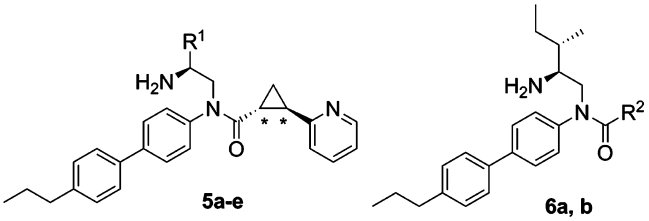
1, 4a-i *(*R*, *R*) and (*S*, *S*) mixture

2 *(*R*, *R*)-isomer

compd[a]	R	pEC$_{50}$ (EC$_{50}$, nM)[b]
2-PCCA (**1**)	Pr	6.04 ± 0.06 (911)
2	Pr	6.22 ± 0.10 (603)
4a	H	5.48 ± 0.08 (3321)
4b	Me	6.07 ± 0.14 (845)
4c	Et	5.70 ± 0.08 (1989)
4d	*i*-Bu	5.74 ± 0.06 (1803)
4e	cyclohexyl	6.13 ± 0.13 (746)
4f	CF_3	5.49 ± 0.08 (3266)
4g	fluoro	NA[c]
4h	chloro	NA[c]
4i	acetyl	6.03 ± 0.05 (923)

[a]All compounds were tested as the HCl salt. [b]pEC$_{50}$ values are means ± standard error of at least three independent experiments performed in duplicate. [c]EC$_{50}$ > 10 μM, tested in two independent experiments performed in duplicate.

Table 2. Structures and Activities of Compounds 5a−e, 6a, and 6b



5a-e **6a, b**

* (*R*, *R*) and (*S*, *S*) mixture

compd[a]	R[1]	R[2]	pEC$_{50}$ (EC$_{50}$, nM)[b]
2-PCCA (**1**)			6.04 ± 0.06 (911)
5a	H		5.27 ± 0.12 (5330)
5b	Me		5.42 ± 0.12 (3821)
5c	*i*-Pr		5.80 ± 0.09 (1602)
5d	*i*-Bu		6.00 ± 0.21 (994)
5e	$PhCH_2$		6.05 ± 0.19 (898)
6a		(1*R*,2*R*)-2-phenyl-cyclopropan-1-yl	5.90 ± 0.05 (1250)
6b		(pyridin-2-yl)methyl	NA[c]

[a]All compounds were tested as the HCl salt. [b]pEC$_{50}$ values are means ± standard error of at least three independent experiments performed in duplicate. [c]EC$_{50}$ > 10 μM

(±)-tert-Butyl (1R,2R*)-2-(pyridin-2-yl)cyclopropanecarboxylate (**9**).* A solution of 2-vinylpyridine (**8**) (0.58 mL, 5.4 mmol), *tert*-butyl diazoacetate (0.88 mL, 6.4 mmol), and 5,10,15,20-tetraphenyl-21*H*,23*H*-porphine cobalt(II) (72 mg, 0.11 mmol) in toluene (25 mL) was heated in a sealed tube at 80 °C for 2 h. After cooling to room temperature, the mixture was concentrated under reduced pressure. Flash column chromatography of the crude product on silica gel using 0−30% EtOAc in hexanes afforded **9** (0.82 g, 69%) as a brown oil: ^{1}H NMR (300 MHz; $CDCl_3$) δ 8.44 (d, *J* = 6.0 Hz, 1H), 7.56 (td, *J* = 6.0, 3.0 Hz, 1H)), 7.23 (t, *J* = 6.0 Hz, 1H), 7.11−6.93 (m, 1H), 2.51 (ddd, *J* = 9.0, 6.0, 3.0 Hz, 1H), 2.16 (ddd, *J* = 9.0, 6.0, 3.0 Hz, 1H), 1.57−1.48 (m, 2H), 1.45 (s, 9H); ^{13}C NMR (75 MHz; $CDCl_3$) δ 172.6, 159.3, 149.4, 135.9, 123.3, 121.1, 80.5, 28.2, 26.8, 25.4, 17.2; MS (ESI) *m*/*z* 220.4 $[M + H]^+$.

(±)-(1R,2R*)-2-(Pyridin-2-yl)cyclopropanecarboxylic Acid Hydrochloride (**10**).* To a solution of **9** (0.80 g, 3.65 mmol) in CH_2Cl_2 (10 mL) was added 4 M HCl in dioxane (3 mL), and the reaction was stirred at room temperature overnight. The solvent was removed under reduced procedure. The residue was triturated with CH_2Cl_2/Et_2O to afford **10** (0.60 g, 81%) as a greenish solid: mp 141−143 °C; ^{1}H NMR (300 MHz; DMSO-d_6) δ 12.00 (br s, 1H), 8.67 (dd, *J* = 6.0, 3.0 Hz, 1H), 8.30 (td, *J* = 6.0, 3.0 Hz, 1H), 7.81−7.68 (m, 2H), 2.90 (ddd, *J* = 9.0, 6.0, 3.0 Hz, 1H), 2.33 (ddd, *J* = 9.0, 6.0, 3.0 Hz, 1H), 1.80−1.60 (m, 2H); ^{13}C NMR (75 MHz; DMSO-d_6) δ 172.5, 156.1, 134.5, 143.0, 123.8, 123.4, 25.0, 23.2, 17.3; MS (ESI) *m*/*z* 164.4 $[M + H]^+$.

*tert-Butyl {(2S,3S)-1-[(4-Bromophenyl)amino]-3-methylpentan-2-yl}carbamate (**13**).* To a solution of (−)-(2*S*,3*S*)-*N*-Boc-2-amino-3-methyl-1-pentanol (2.17 g, 10.0 mmol) in water-saturated CH_2Cl_2 (10 mL) at room temperature was added Dess-Martin reagent (8.90 g, 21.0 mmol), and the reaction was stirred for 1 h. Additional water-saturated CH_2Cl_2 (5 mL) was added every 15 min during the reaction time. The mixture was diluted with Et_2O (100 mL) and poured into a solution of $Na_2S_2O_3$ (17 g) in 80% saturated $NaHCO_3$ (100 mL). After stirring for 10 min, the layers were separated and the aqueous layer was extracted with Et_2O (100 mL). The combined organic layers were washed with ice-cold saturated $NaHCO_3$ (30 mL) and water (30 mL). The solution was dried (Na_2SO_4) and concentrated under reduced pressure to give the crude aldehyde **12**. To a solution of 4-bromoaniline (**11**) (1.72 g, 10.0 mmol) in dichloroethane (60 mL) was added the above crude aldehyde, followed by $NaBH(OAc)_3$ (4.24 g, 20.0 mmol). The mixture was stirred at room temperature overnight. Saturated $NaHCO_3$ (20 mL) was added, and the layers were separated. The aqueous layer was extracted with CH_2Cl_2 (2 × 30 mL). The combined organic layers were washed with brine (3 × 30 mL), dried (Na_2SO_4) and concentrated under reduced pressure. Flash column chromatography of the crude product on silica gel using 0−30% EtOAc in hexanes afforded **13** (2.78 g, 75%) as a white solid: mp 103−105 °C; $[\alpha]^{23}_D$ +11.4° (*c* 0.52, CH_3OH); ^{1}H NMR (300 MHz; $CDCl_3$) δ 7.24 (d, *J* = 9.0 Hz, 2H), 6.45 (d, *J* = 9.0 Hz, 2H), 4.52 (d, *J* = 9.0 Hz, 1H), 4.20 (br s, 1H), 3.82−3.65 (m, 1H), 3.30−3.14 (m, 1H), 3.05−2.89 (m, 1H), 1.60−1.47 (m, 1H), 1.44 (s, 9H), 1.23−1.10 (m, 1H), 0.95 (d, *J* = 6.0 Hz, 3H), 0.95 (t, *J* = 7.5 Hz, 3H); ^{13}C NMR (75 MHz; $CDCl_3$) δ 154.9, 145.7, 130.1, 112.4, 106.9, 77.9, 52.9, 45.0, 35.7, 26.6, 23.6, 13.8, 9.9; MS (ESI) *m*/*z* 371.3 $[M + H]^+$ (^{79}Br), 373.3 $[M + H]^+$ (^{81}Br).

tert-Butyl [(2S,3S)-1-{4-Bromophenyl-[(1R,2R*)-2-(pyridin-2-yl)-cyclopropanecarbonyl]amino}-3-methylpentan-2-yl]carbamate (**14**).* To a solution of **10** (0.40 g, 2.0 mmol) in CH_2Cl_2 (20 mL) at room temperature was added oxalyl chloride (0.35 mL, 4.0 mmol) and DMF (50 μL). The mixture was stirred at 40 °C for 2 h and then cooled to room temperature and concentrated under reduced pressure. The residue was dissolved in CH_2Cl_2 (20 mL) and treated with **13** (0.74 g, 2.0 mmol) and Et_3N (1.1 mL, 8.0 mmol). The resulting solution was stirred at room temperature overnight. Saturated $NaHCO_3$ (10 mL) was added, and the layers were separated. The aqueous layer was extracted with CH_2Cl_2 (3 × 10 mL). The combined organic layers were washed with brine (3 × 20 mL), dried (Na_2SO_4) and concentrated under reduced pressure. Flash column chromatography of the crude product on silica gel using 0−25% EtOAc in hexanes afforded **14** (0.74 g, 72%, 1:1 diastereomeric mixture) as a light yellow foam: ^{1}H NMR (300 MHz; $CDCl_3$) δ 8.30 (d, *J* = 6.0 Hz, 1H), 7.58−7.34 (m, 3H), 7.22−6.98 (m, 4H), 4.96 (d, *J* = 9.0 Hz, 1H), 4.45−4.25 (m, 1H), 3.80−3.78 (m, 1H), 3.21−3.06 (m, 1H), 2.71−2.62 (m, 0.5 H), 2.58−2.48 (m, 0.5 H), 1.98−1.86 (m, 1H), 1.75−1.62 (m, 1H), 1.60−1.35 (m, 3H), 1.45 and 1.40 (2s, 9H), 1.15−1.02 (m, 1H), 0.92−0.80 (m, 6H); MS (ESI) *m*/*z* 516.7 $[M + H]^+$ (^{79}Br), 518.6 $[M + H]^+$ (^{81}Br).

dx.doi.org/10.1021/cn500082p | ACS Chem. Neurosci. 2014, 5, 576−587

tert-Butyl [(2S,3S)-1-{(4′-Propylbiphenyl-4-yl)-[(1R,2R*)-2-(pyridin-2-yl)cyclopropanecarbonyl]amino}-3-methylpentan-2-yl]carbamate (**15a**).* A mixture of **14** (0.48 g, 0.92 mmol), 4-propylphenylboronic acid (0.25 g, 1.4 mmol), $Pd(dppf)Cl_2·CH_2Cl_2$ (70 mg, 0.092 mmol), and K_3PO_4 (0.58 g, 2.7 mmol) in dimethoxyethane (10.5 mL) and water (3.5 mL) was heated in a sealed vessel by microwave irradiation at 160 °C for 6 min. The resulting mixture was poured into 1 N NaOH solution (30 mL) and extracted with CH_2Cl_2 (3 × 30 mL). The combined organic layers were dried (Na_2SO_4) and concentrated under reduced pressure. Flash column chromatography of the crude product on silica gel using 0−20% EtOAc in hexanes afforded **15a** (0.49 g, 96%, 1:1 diastereomeric mixture) as a white foam: ^{1}H NMR (300 MHz; $CDCl_3$) δ 8.28−8.22 (m, 1H), 7.58−7.38 (m, 5H), 7.32−7.13 (m, 5H), 7.03−6.96 (m, 1H), 5.12−5.05 (m, 1H), 4.48−4.36 (m, 1H), 3.85−3.56 (m, 1H), 3.24−3.13 (m, 1H), 2.71−2.50 (m, 3H), 2.05−1.95 (m, 1H), 1.75−1.58 (m, 3H), 1.51−1.38 (m, 3H), 1.47 and 1.43 (2s, 9H), 1.56−1.40 (m, 1H), 0.97 (t, J = 7.5 Hz, 3H), 0.92−0.80 (m, 6H); MS (ESI) m/z 557.0 $[M + H]^+$.

tert-Butyl [(2S,3S)-1-{(Biphenyl-4-yl)-[(1R,2R*)-2-(pyridin-2-yl)-cyclopropanecarbonyl]amino}-3-methylpentan-2-yl]carbamate (**15b**).* The procedure for **15a** was followed using 30 mg (0.058 mmol) of **14** and 11 mg (0.087 mmol) of phenylboronic acid to give 24 mg (81%) of **15b** as a 1:1 diastereomeric mixture: ^{1}H NMR (300 MHz; $CDCl_3$) δ 8.28−8.22 (m, 1H), 7.59−7.38 (m, 6H), 7.37−7.14 (m, 5H), 7.12−6.95 (m, 1H), 5.14−5.07 (m, 1H), 4.49−4.36 (m, 1H), 3.85−3.68 (m, 1H), 3.26−3.14 (m, 1H), 2.75−2.66 (m, 0.5 H), 2.60−2.50 (m, 0.5H), 2.08−1.98 (m, 1H), 1.75−1.66 (m, 0.5H), 1.65−1.57 (m, 0.5H), 1.55−1.36 (m, 3H), 1.47 and 1.43 (2s, 9H), 1.18−1.02 (m, 1H), 0.94−0.80 (m, 6H); MS (ESI) m/z 514.7 $[M + H]^+$.

tert-Butyl [(2S,3S)-1-{(4′-Methylbiphenyl-4-yl)-[(1R,2R*)-2-(pyridin-2-yl)cyclopropanecarbonyl]amino}-3-methylpentan-2-yl]carbamate (**15c**).* The procedure for **15a** was followed using 30 mg (0.058 mmol) of **14** and 12 mg (0.087 mmol) of 4-methylphenylboronic acid to give 20 mg (65%) of **15c** as a 1:1 diastereomeric mixture: ^{1}H NMR (300 MHz; $CDCl_3$) δ 8.29−8.22 (m, 1H), 7.58−7.38 (m, 5H), 7.37−7.16 (m, 5H), 7.02−6.92 (m, 1H), 5.13−5.06 (m, 1H), 4.48−4.37 (m, 1H), 3.83−3.68 (m, 1H), 3.24−3.12 (m, 1H), 2.73−2.65 (m, 0.5 H), 2.60−2.50 (m, 0.5H), 2.39 (s, 3H), 2.08−1.96 (m, 1H), 1.76−1.55 (m, 1H), 1.54−1.37 (m, 3H), 1.47 and 1.43 (2s, 9H), 1.17−1.00 (m, 1H), 0.92−0.78 (m, 6H); MS (ESI) m/z 528.7 $[M + H]^+$.

tert-Butyl [(2S,3S)-1-{(4′-Ethylbiphenyl-4-yl)-[(1R,2R*)-2-(pyridin-2-yl)cyclopropanecarbonyl]amino}-3-methylpentan-2-yl]carbamate (**15d**).* The procedure for **15a** was followed using 30 mg (0.058 mmol) of **14** and 13 mg (0.087 mmol) of 4-ethylphenylboronic acid to give 25 mg (80%) of **15d** as a 1:1 diastereomeric mixture: ^{1}H NMR (300 MHz; $CDCl_3$) δ 8.28−8.23 (m, 1H), 7.58−7.38 (m, 5H), 7.37−7.15 (m, 5H), 7.04−6.95 (m, 1H), 5.15−5.05 (m, 1H), 4.50−4.37 (m, 1H), 3.85−3.68 (m, 1H), 3.25−3.14 (m, 1H), 2.76−2.66 (m, 2.5 H), 2.60−2.48 (m, 0.5H), 2.10−1.96 (m, 1H), 1.76−1.54 (m, 1H), 1.53−1.37 (m, 3H), 1.47 and 1.43 (2s, 9H), 1.28 (t, J = 7.5 Hz, 3H), 1.17−1.00 (m, 1H), 0.93−0.78 (m, 6H); MS (ESI) m/z 542.6 $[M + H]^+$.

tert-Butyl [(2S,3S)-1-{(4′-Isobutylbiphenyl-4-yl)-[(1R,2R*)-2-(pyridin-2-yl)cyclopropanecarbonyl]amino}-3-methylpentan-2-yl]carbamate (**15e**).* The procedure for **15a** was followed using 30 mg (0.058 mmol) of **14** and 16 mg (0.087 mmol) of 4-isobutylphenylboronic acid to give 23 mg (77%) of **15e** as a 1:1 diastereomeric mixture: ^{1}H NMR (300 MHz; $CDCl_3$) δ 8.28−8.23 (m, 1H), 7.60−7.38 (m, 5H), 7.34−7.15 (m, 5H), 7.04−6.95 (m, 1H), 5.14−5.05 (m, 1H), 4.50−4.36 (m, 1H), 3.86−3.68 (m, 1H), 3.24−3.14 (m, 1H), 2.74−2.65 (m, 0.5 H), 2.60−2.46 (m, 2.5H), 2.10−1.82 (m, 2H), 1.76−1.54 (m, 1H), 1.53−1.36 (m, 3H), 1.47 and 1.43 (2s, 9H), 1.16−1.00 (m, 1H), 0.98−0.78 (m, 12H); MS (ESI) m/z 570.6 $[M + H]^+$.

tert-Butyl [(2S,3S)-1-{(4′-Cyclohexylbiphenyl-4-yl)-[(1R,2R*)-2-(pyridin-2-yl)cyclopropanecarbonyl]amino}-3-methylpentan-2-yl]carbamate (**15f**).* The procedure for **15a** was followed using 30 mg (0.058 mmol) of **14** and 18 mg (0.087 mmol) of 4-cyclohexylphenylboronic acid to give 28 mg (81%) of **15f** as a 1:1 diastereomeric mixture: ^{1}H NMR (300 MHz; $CDCl_3$) δ 8.28−8.21 (m, 1H), 7.58−7.40 (m, 5H), 7.32−7.12 (m, 5H), 7.02−6.92 (m, 1H), 5.15−5.05 (m, 1H), 4.50−4.36 (m, 1H), 3.88−3.68 (m, 1H), 3.26−3.13 (m, 1H), 2.75−2.64 (m, 0.5 H), 2.60−2.48 (m, 1.5H), 2.05−1.71 (m, 7H), 1.69−1.25 (m, 8H), 1.47 and 1.43 (2s, 9H), 1.17−1.00 (m, 1H), 0.96−0.80 (m, 6H); MS (ESI) m/z 596.9 $[M + H]^+$.

tert-Butyl [(2S,3S)-1-{(4′-Trifluoromethylbiphenyl-4-yl)-[(1R,2R*)-2-(pyridin-2-yl)cyclopropanecarbonyl]amino}-3-methylpentan-2-yl]carbamate (**15g**).* The procedure for **15a** was followed using 30 mg (0.058 mmol) of **14** and 17 mg (0.087 mmol) of 4-trifluoromethylphenylboronic acid to give 25 mg (74%) of **15g** as a 1:1 diastereomeric mixture: ^{1}H NMR (300 MHz; $CDCl_3$) δ 8.30−8.22 (m, 1H), 7.72−7.40 (m, 7H), 7.38−7.16 (m, 3H), 7.02−6.93 (m, 1H), 5.13−5.02 (m, 1H), 4.50−4.38 (m, 1H), 3.84−3.65 (m, 1H), 3.28−3.15 (m, 1H), 2.76−2.65 (m, 0.5 H), 2.61−2.48 (m, 0.5H), 2.08−1.96 (m, 1H), 1.76−1.55 (m, 1H), 1.54−1.37 (m, 3H), 1.47 and 1.43 (2s, 9H), 1.18−1.00 (m, 1H), 0.94−0.78 (m, 6H); MS (ESI) m/z 582.7 $[M + H]^+$.

tert-Butyl [(2S,3S)-1-{(4′-Fluorobiphenyl-4-yl)-[(1R,2R*)-2-(pyridin-2-yl)cyclopropanecarbonyl]amino}-3-methylpentan-2-yl]carbamate (**15h**).* The procedure for **15a** was followed using 30 mg (0.058 mmol) of **14** and 12 mg (0.087 mmol) of 4-fluorophenylboronic acid to give 20 mg (74%) of **15h** as a 1:1 diastereomeric mixture: ^{1}H NMR (300 MHz; $CDCl_3$) δ 8.30−8.22 (m, 1H), 7.56−7.48 (m, 4H), 7.30−7.18 (m, 6H), 7.02−7.92 (m, 1H), 5.12−5.05 (m, 1H), 4.52−4.38 (m, 1H), 3.82−3.65 (m, 1H), 3.25−3.12 (m, 1H), 2.76−2.65 (m, 0.5 H), 2.61−2.48 (m, 0.5H), 2.08−1.95 (m, 1H), 1.78−1.55 (m, 1H), 1.54−1.37 (m, 3H), 1.47 and 1.43 (2s, 9H), 1.18−1.00 (m, 1H), 0.95−0.80 (m, 6H); MS (ESI) m/z 532.5 $[M + H]^+$.

tert-Butyl [(2S,3S)-1-{(4′-Chlorobiphenyl-4-yl)-[(1R,2R*)-2-(pyridin-2-yl)cyclopropanecarbonyl]amino}-3-methylpentan-2-yl]carbamate (**15i**).* The procedure for **15a** was followed using 30 mg (0.058 mmol) of **14** and 14 mg (0.087 mmol) of 4-chlorophenylboronic acid to give 22 mg (74%) of **15i** as a 1:1 diastereomeric mixture: ^{1}H NMR (300 MHz; $CDCl_3$) δ 8.30−8.21 (m, 1H), 7.55−7.36 (m, 6H), 7.30−7.15 (m, 4H), 7.02−7.95 (m, 1H), 5.10−5.00 (m, 1H), 4.50−4.35 (m, 1H), 3.80−3.62 (m, 1H), 3.26−3.15 (m, 1H), 2.76−2.65 (m, 0.5 H), 2.61−2.48 (m, 0.5H), 2.08−1.95 (m, 1H), 1.75−1.55 (m, 1H), 1.54−1.37 (m, 3H), 1.47 and 1.42 (2s, 9H), 1.16−1.00 (m, 1H), 0.95−0.80 (m, 6H); MS (ESI) m/z 548.5 $[M + H]^+$.

tert-Butyl [(2S,3S)-1-{(4′-Acetylbiphenyl-4-yl)-[(1R,2R*)-2-(pyridin-2-yl)cyclopropanecarbonyl]amino}-3-methylpentan-2-yl]carbamate (**15j**).* The procedure for **15a** was followed using 30 mg (0.058 mmol) of **14** and 14 mg (0.087 mmol) of 4-acetylphenylboronic acid to give 25 mg (76%) of **15j** as a 1:1 diastereomeric mixture: ^{1}H NMR (300 MHz; $CDCl_3$) δ 8.30−8.21 (m, 1H), 8.06−7.98 (m, 2H), 7.68−7.48 (m, 5H), 7.38−7.15 (m, 3H), 7.02−7.93 (m, 1H), 5.10−5.00 (m, 1H), 4.50−4.36 (m, 1H), 3.80−3.62 (m, 1H), 3.28−3.15 (m, 1H), 2.75−2.65 (m, 0.5 H), 2.64 (s, 3H), 2.61−2.48 (m, 0.5H), 2.08−1.95 (m, 1H), 1.76−1.55 (m, 1H), 1.54−1.37 (m, 3H), 1.47 and 1.43 (2s, 9H), 1.16−0.98 (m, 1H), 0.92−0.78 (m, 6H); MS (ESI) m/z 557.2 $[M + H]^+$.

(1R,2R*)-2-(Pyridin-2-yl)cyclopropanecarboxylic Acid [(2S,3S)-2-Amino-3-methylpentyl]-(4′-propylbiphenyl-4-yl)amide Dihydrochloride (**1**).* A solution of **15a** (150 mg, 0.27 mmol) and 4 M HCl in dioxane (2 mL) in CH_2Cl_2 (5 mL) was stirred at room temperature for 6 h. The solvent was removed under reduced pressure. The resulting residue was triturated with Et_2O to give **1** (135 mg, 95%, 1:1 diastereomeric mixture) as a white solid: ^{1}H NMR (300 MHz; CD_3OD) δ 8.66−8.54 (m, 1H), 8.37−8.23 (m, 1H), 7.82−7.44 (m, 8H), 7.32−7.22 (m, 2H), 4.43 (dd, J = 15.0, 9.0 Hz, 0.5H), 4.28 (dd, J = 15.0, 9.0 Hz, 0.5H), 3.82 (d, J = 15.0 Hz, 0.5H), 3.68 (d, J = 15.0 Hz, 0.5H), 3.46−3.35 (m, 1H), 3.11−3.02 (m, 0.5H), 3.00−2.90 (m, 0.5H), 2.62 (t, J = 7.5 Hz, 2H), 2.22−2.08 (m, 1H), 2.02−1.88 (m, 1H), 1.87−1.62 (m, 4H), 1.50−1.12 (m, 2H), 1.02−0.78 (m, 9H); HRMS (ESI) calcd. for $C_{30}H_{37}N_3O$ $[M + H]^+$: 456.3009. Found: 456.3020.

(1R,2R*)-2-(Pyridin-2-yl)cyclopropanecarboxylic Acid [(2S,3S)-2-Amino-3-methylpentyl]-(biphenyl-4-yl)amide Dihydrochloride (**4a**).* The procedure for **1** was followed using 20 mg (0.039 mmol) of **15b** and 1 mL of 4 M HCl in dioxane to give 17 mg (90%) of **4a** as a 1:1 diastereomeric mixture: ^{1}H NMR (300 MHz; CD_3OD) δ 8.28−8.21

(m, 1H), 7.72−7.52 (m, 5H), 7.48−7.32 (m, 5H), 7.30−7.23 (m, 1H), 7.19−7.10 (m, 1H), 4.40−4.22 (m, 1H), 3.80−3.66 (m, 1H), 3.40−3.22 (m, 1H), 2.70−2.53 (m, 1H), 2.05−1.90 (m, 1H), 1.84−1.62 (m, 2H), 1.50−1.15 (m, 3H), 0.99 and 0.97 (2d, J = 6.0 Hz, 3H), 0.90−0.80 (m, 3H); HRMS (ESI) calcd. for $C_{27}H_{31}N_3O$ [M + H]$^+$: 414.2541. Found: 414.2540.

(1R,2R*)-2-(Pyridin-2-yl)cyclopropanecarboxylic Acid [(2S,3S)-2-Amino-3-methylpentyl]-(4′-methylbiphenyl-4-yl)amide Dihydrochloride (**4b**).* The procedure for **1** was followed using 20 mg (0.038 mmol) of **15c** and 1 mL of 4 M HCl in dioxane to give 18 mg (95%) of **4b** as a 1:1 diastereomeric mixture: ^{1}H NMR (300 MHz; CD_3OD) δ 8.68−8.55 (m, 1H), 8.42−8.30 (m, 1H), 7.88−7.40 (m, 8H), 7.30−7.20 (m, 2H), 4.50−4.40 (m, 0.5H), 4.40−4.28 (m, 0.5H), 3.88−3.56 (m, 1H), 3.48−3.38 (m, 1H), 3.12−3.05 (m, 0.5H), 3.05−2.96 (m, 0.5H), 2.37 (s, 3H), 2.22−2.10 (m, 1H), 2.10−1.88 (m, 1H), 1.87−1.60 (m, 2H), 1.48−1.15 (m, 2H), 1.06−0.80 (m, 6H); HRMS (ESI) calcd. for $C_{28}H_{33}N_3O$ [M + H]$^+$: 428.2696. Found: 428.2701.

(1R,2R*)-2-(Pyridin-2-yl)cyclopropanecarboxylic Acid [(2S,3S)-2-Amino-3-methylpentyl]-(4′-ethylbiphenyl-4-yl)amide Dihydrochloride (**4c**).* The procedure for **1** was followed using 20 mg (0.037 mmol) of **15d** and 1 mL of 4 M HCl in dioxane to give 18 mg (95%) of **4c** as a 1:1 diastereomeric mixture: ^{1}H NMR (300 MHz; CD_3OD) δ 8.70−8.58 (m, 1H), 8.45−8.30 (m, 1H), 7.88−7.45 (m, 8H), 7.32−7.20 (m, 2H), 4.52−4.40 (m, 0.5H), 4.38−4.28 (m, 0.5H), 3.88−3.56 (m, 1H), 3.48−3.35 (m, 1H), 3.15−3.05 (m, 0.5H), 3.05−2.95 (m, 0.5H), 2.80−2.60 (m, 2H), 2.25−2.10 (m, 1H), 2.10−1.90 (m, 1H), 1.87−1.60 (m, 2H), 1.50−1.10 (m, 5H), 1.08−0.80 (m, 6H); HRMS (ESI) calcd. for $C_{29}H_{35}N_3O$ [M + H]$^+$: 442.2853. Found: 442.2867.

(1R,2R*)-2-(Pyridin-2-yl)cyclopropanecarboxylic Acid [(2S,3S)-2-Amino-3-methylpentyl]-(4′-isobutylbiphenyl-4-yl)amide Dihydrochloride (**4d**).* The procedure for **1** was followed using 20 mg (0.035 mmol) of **15e** and 1 mL of 4 M HCl in dioxane to give 17 mg (90%) of **4d** as a 1:1 diastereomeric mixture: ^{1}H NMR (300 MHz; CD_3OD) δ 8.68−8.58 (m, 1H), 8.40−8.28 (m, 1H), 7.92−7.43 (m, 8H), 7.30−7.20 (m, 2H), 4.50−4.40 (m, 0.5H), 4.38−4.25 (m, 0.5H), 3.88−3.60 (m, 1H), 3.50−3.38 (m, 1H), 3.13−3.05 (m, 0.5H), 3.05−2.90 (m, 0.5H), 2.51 (d, J = 6.0 Hz, 2H), 2.25−2.10 (m, 1H), 2.05−1.60 (m, 4H), 1.50−1.20 (m, 2H), 1.10−0.76 (m, 12H); HRMS (ESI) calcd. for $C_{31}H_{39}N_3O$ [M + H]$^+$: 470.3166. Found: 470.3178.

(1R,2R*)-2-(Pyridin-2-yl)cyclopropanecarboxylic Acid [(2S,3S)-2-Amino-3-methylpentyl]-(4′-cyclohexylbiphenyl-4-yl)amide Dihydrochloride (**4e**).* The procedure for **1** was followed using 20 mg (0.034 mmol) of **15f** and 1 mL of 4 M HCl in dioxane to give 19 mg (98%) of **4e** as a 1:1 diastereomeric mixture: ^{1}H NMR (300 MHz; CD_3OD) δ 8.55−8.40 (m, 1H), 8.15−8.00 (m, 1H), 7.75−7.40 (m, 8H), 7.35−7.20 (m, 2H), 4.48−4.20 (m, 1H), 3.88−3.68 (m, 1H), 3.42−3.35 (m, 1H), 2.98−2.88 (m, 1H), 2.68−2.48 (m, 1H), 2.20−2.00 (m, 1H), 1.98−1.70 (m, 7H), 1.66−1.10 (m, 8H), 1.05−0.78 (m, 6H); HRMS (ESI) calcd. for $C_{33}H_{41}N_3O$ [M + H]$^+$: 496.3322. Found: 496.3323.

(1R,2R*)-2-(Pyridin-2-yl)cyclopropanecarboxylic Acid [(2S,3S)-2-Amino-3-methylpentyl]-(4′-trifluoromethylbiphenyl-4-yl)amide Dihydrochloride (**4f**).* The procedure for **1** was followed using 20 mg (0.034 mmol) of **15g** and 1 mL of 4 N HCl in dioxane to give 18 mg (95%) of **4f** as a 1:1 diastereomeric mixture: ^{1}H NMR (300 MHz; CD_3OD) δ 8.70−8.58 (m, 1H), 8.42−8.30 (m, 1H), 7.90−7.50 (m, 10H), 4.55−4.40 (m, 0.5H), 4.40−4.26 (m, 0.5H), 3.90−3.60 (m, 1H), 3.50−3.38 (m, 1H), 3.15−3.05 (m, 0.5H), 3.05−2.92 (m, 0.5H), 2.25−2.10 (m, 1H), 2.10−1.92 (m, 1H), 1.92−1.62 (m, 2H), 1.50−1.15 (m, 2H), 1.08−0.80 (m, 6H); HRMS (ESI) calcd. for $C_{28}H_{30}F_3N_3O$ [M + H]$^+$: 482.2414. Found: 482.2416.

(1R,2R*)-2-(Pyridin-2-yl)cyclopropanecarboxylic Acid [(2S,3S)-2-Amino-3-methylpentyl]-(4′-fluorobiphenyl-4-yl)amide Dihydrochloride (**4g**).* The procedure for **1** was followed using 20 mg (0.038 mmol) of **15h** and 1 mL of 4 M HCl in dioxane to give 18 mg (94%) of **4g** as a 1:1 diastereomeric mixture: ^{1}H NMR (300 MHz; CD_3OD) δ 8.70−8.58 (m, 1H), 8.45−8.35 (m, 1H), 7.90−7.50 (m, 8H), 7.25−7.10 (m, 2H), 4.52−4.40 (m, 0.5H), 4.40−4.25 (m, 0.5H), 3.90−3.60 (m, 1H), 3.52−3.40 (m, 1H), 3.16−3.06 (m, 0.5H), 3.05−2.92 (m, 0.5H), 2.25−2.10 (m, 1H), 2.10−1.92 (m, 1H), 1.92−1.60 (m, 2H), 1.50−1.18 (m, 2H), 1.10−0.80 (m, 6H); HRMS (ESI) calcd. for $C_{27}H_{30}FN_3O$ [M + H]$^+$: 432.2446. Found: 432.2453.

(1R,2R*)-2-(Pyridin-2-yl)cyclopropanecarboxylic Acid [(2S,3S)-2-Amino-3-methylpentyl]-(4′-chlorobiphenyl-4-yl)amide Dihydrochloride (**4h**).* The procedure for **1** was followed using 20 mg (0.036 mmol) of **15i** and 1 mL of 4 M HCl in dioxane to give 18 mg (96%) of **4h** as a 1:1 diastereomeric mixture: ^{1}H NMR (300 MHz; CD_3OD) δ 8.72−8.58 (m, 1H), 8.45−8.35 (m, 1H), 7.90−7.55 (m, 8H), 7.55−7.40 (m, 2H), 4.55−4.42 (m, 0.5H), 4.40−4.25 (m, 0.5H), 3.90−3.60 (m, 1H), 3.50−3.40 (m, 1H), 3.18−3.07 (m, 0.5H), 3.06−2.96 (m, 0.5H), 2.25−2.10 (m, 1H), 2.08−1.90 (m, 1H), 1.90−1.68 (m, 2H), 1.58−1.12 (m, 2H), 1.10−0.80 (m, 6H); HRMS (ESI) calcd. for $C_{27}H_{30}ClN_3O$ [M + H]$^+$: 448.2150. Found: 448.2162.

(1R,2R*)-2-(Pyridin-2-yl)cyclopropanecarboxylic Acid [(2S,3S)-2-Amino-3-methylpentyl]-(4′-acetylbiphenyl-4-yl)amide Dihydrochloride (**4i**).* The procedure for **1** was followed using 20 mg (0.036 mmol) of **15j** and 1 mL of 4 M HCl in dioxane to give 17 mg (90%) of **4i** as a 1:1 diastereomeric mixture: ^{1}H NMR (300 MHz; CD_3OD) δ 8.35−8.22 (m, 1H), 8.05−7.92 (m, 2H), 7.90−7.75 (m, 1H), 7.72−7.60 (m, 4H), 7.52−7.40 (m, 2H), 7.35−7.22 (m, 2H), 4.38−4.16 (m, 1H), 3.80−3.55 (m, 1H), 3.38−3.20 (m, 1H), 2.80−2.60 (m, 1H), 2.54 (s, 3H), 2.00−1.88 (m, 1H), 1.78−1.60 (m, 2H), 1.50−1.10 (m, 3H), 0.95−0.70 (m, 6H); HRMS (ESI) calcd. for $C_{29}H_{33}N_3O_2$ [M + H]$^+$: 456.2646. Found: 456.2658.

*tert-Butyl (1R,2R)-2-(Pyridin-2-yl)cyclopropanecarboxylate ((1R,2R)-**9**).* An oven-dried Schlenk tube, that was previously charged with chiral porphyrin catalyst [Co(3,5-Di^{t}Bu-ChenPhyrin)][11] (3.4 mg, 0.0025 mmol) and DMAP (15 mg, 0.13 mmol), was evacuated and backfilled with nitrogen gas. Toluene (0.5 mL) and 2-vinylpyridine (**8**) (26 mg, 0.25 mmol) were added, followed by the remaining solvent (total 1.0 mL). The Schlenk tube was then placed in a −20 °C cooling bath, and *tert*-butyl diazoacetate (41 μL, 0.3 mmol) was added dropwise. After addition, the tube was purged with nitrogen for 2 min and the mixture was stirred at −20 °C for 12 h, then at 0 °C for 12 h. After warming to room temperature, the reaction was continued for another 24 h. Flash column chromatography of the crude mixture on silica gel using 10% EtOAc in hexanes afforded (1*R*,2*R*)-**9** (35 mg, 64%) as a colorless oil. Assignment of the absolute configuration was made based on the analogy to similar compounds with known absolute configurations synthesized using the same chiral catalyst.[11] The enantiomeric excess (97% ee) was determined by HPLC (ChiralPak AD-H column; 0.5% isopropanol/hexanes; flow rate 0.8 mL/min; detection 254 nm; retention time 10.6 min).

*(1R,2R)-2-(Pyridin-2-yl)cyclopropanecarboxylic Acid Hydrochloride ((1R,2R)-**10**).* To a solution of (1*R*,2*R*)-**9** (110 mg, 0.50 mmol) in CH_2Cl_2 (5 mL) was added 4 M HCl in dioxane (2 mL), and the reaction was stirred at room temperature for 5 h. The solvent was removed under reduced procedure. The residue was triturated with CH_2Cl_2/Et_2O to afford (1*R*,2*R*)-**10** (95 mg, 95%) as a white solid: mp 155−157 °C; $[\alpha]^{23}_D$ −206.8° (c 0.5, CH_3OH).

*tert-Butyl [(2S,3S)-1-{4-Bromophenyl-[(1R,2R)-2-(pyridin-2-yl)-cyclopropanecarbonyl]amino}-3-methylpentan-2-yl]carbamate ((1R,2R)-**14**).* The procedure for **14** was followed using 108 mg (0.29 mmol) of **13** and 70 mg (0.35 mmol) of (1*R*,2*R*)-**10** to give 97 mg (65%) of (1*R*,2*R*)-**14** as a white foam: $[\alpha]^{23}_D$ −21.0° (c 0.58, CH_3OH); ^{1}H NMR (300 MHz; $CDCl_3$) δ 8.30 (d, J = 6.0 Hz, 1H), 7.56−7.40 (m, 3H), 7.22−7.06 (m, 3H), 7.06−6.98 (m, 1H), 4.96 (d, J = 9.0 Hz, 1H), 4.36 (t, J = 12.0 Hz, 1H), 3.78−3.65 (m, 1H), 3.12 (dd, J = 12.0, 3.0 Hz, 1H), 2.67 (ddd, J = 9.0, 6.0, 3.0 Hz, 1H), 1.93−1.86 (m, 1H), 1.61−1.53 (m, 1H), 1.52−1.41 (m, 3H), 1.45 (s, 9H), 1.15−1.02 (m, 1H), 0.87 (d, J = 6.0 Hz, 3H), 0.85 (t, J = 7.5 Hz, 3H); ^{13}C NMR (75 MHz; $CDCl_3$) δ 173.2, 159.3, 156.4, 149.4, 141.3, 136.0, 133.1, 130.1, 122.7, 121.9, 121.2, 79.0, 54.0, 50.3, 38.3, 28.6, 27.5, 25.3, 24.7, 18.3, 15.2, 11.9; MS (ESI) m/z 516.5 [M + H]$^+$ (^{79}Br), 518.4 [M + H]$^+$ (^{81}Br). The diastereomeric excess (>97% de) was determined by HPLC (ChiralPak IA column; 5% EtOH/hexanes; flow rate 1 mL/min; detection 220 nm; retention time 9.1 min).

*tert-Butyl [(2S,3S)-1-{4-Bromophenyl-[(1S,2S)-2-(pyridin-2-yl)-cyclopropanecarbonyl]amino}-3-methylpentan-2-yl]carbamate ((1S,2S)-**14**).* The diastereomeric mixture **14** (350 mg) was separated to (1*R*,2*R*)-**14** (140 mg) and (1*S*,2*S*)-**14** (137 mg) by preparative HPLC using ChiralPak IA column: mobile phase, 5% EtOH/hexanes; flow rate, 5 mL/min; detection 220 nm. The diastereomeric excess

(de) of both of separated compounds was determined to be >99% by HPLC (ChiralPak IA column; 5% EtOH/hexanes; flow rate 1 mL/min; detection 220 nm; retention time, (1*R*,2*R*)-**14**: 9.1 min, (1*S*,2*S*)-**14**: 11.5 min). (1*S*,2*S*)-**14**: White foam; $[\alpha]^{23}_{D}$ +52.7° (*c* 0.55, CH_3OH); ^1H NMR (300 MHz; $CDCl_3$) δ 8.30 (d, *J* = 6.0 Hz, 1H), 7.52 (td, *J* = 9.0, 3.0 Hz, 1H), 7.40 (d, *J* = 6.0 Hz, 2H), 7.16 (d, *J* = 9.0 Hz, 1H), 7.12−6.99 (m, 3H), 4.96 (d, *J* = 9.0 Hz, 1H), 4.38 (dd, *J* = 15.0, 12.0 Hz, 1H), 3.72−3.58 (m, 1H), 3.15 (dd, *J* = 15.0, 3.0 Hz, 1H), 2.52 (ddd, *J* = 9.0, 6.0, 3.0 Hz, 1H), 1.97−1.90 (m, 1H), 1.70−1.63 (m, 1H), 1.54−1.38 (m, 3H), 1.40 (s, 9H), 1.14−1.02 (m, 1H), 0.87 (d, *J* = 6.0 Hz, 3H), 0.84 (t, *J* = 7.5 Hz, 3H); ^{13}C NMR (75 MHz; $CDCl_3$) δ 173.1, 159.2, 156.2, 149.5, 141.1, 135.9, 132.9, 130.1, 122.5, 121.8, 121.2, 78.9, 53.8, 50.0, 38.2, 28.6, 28.1, 25.3, 24.7, 17.7, 15.2, 11.9; MS (ESI) *m*/*z* 516.5 $[M + H]^+$ (^{79}Br), 518.4 $[M + H]^+$ (^{81}Br).

*(1R,2R)-2-(Pyridin-2-yl)cyclopropanecarboxylic Acid [(2S,3S)-2-Amino-3-methylpentyl]-(4′-propylbiphenyl-4-yl)amide Dihydrochloride (**2**).* The procedure for **15a** was followed using 120 mg (0.23 mmol) of (1*R*,2*R*)-**14**, followed by deprotection of the Boc protecting group with 4 M HCl in dioxane, to give 97 mg (80% over two steps) of **2** as a white solid: mp 125 °C (fusion); $[\alpha]^{23}_{D}$ +6.3° (*c* 1, CH_3OH); ^1H NMR (300 MHz; CD_3OD) δ 8.58 (d, *J* = 6.0 Hz, 1H), 8.28 (t, *J* = 9.0 Hz, 1H), 7.78−7.52 (m, 6H), 7.51 (d, *J* = 9.0 Hz, 2H), 7.27 (d, *J* = 9.0 Hz, 2H), 4.29 (dd, *J* = 15.0, 9.0 Hz, 1H), 3.83 (d, *J* = 12.0 Hz, 1H), 3.48−3.38 (m, 1H), 3.01−2.90 (m, 1H), 2.62 (t, *J* = 7.5 Hz, 2H), 2.22−2.12 (m, 1H), 2.02−1.92 (m, 1H), 1.90−1.75 (M, 1H), 1.75−1.60 (m, 3H), 1.50−1.35 (m, 1H), 1.35−1.15 (m, 1H), 1.05−0.90 (m, 6H), 0.86 (t, *J* = 6.0 Hz, 3H); ^{13}C NMR (75 MHz; CD_3OD) δ 173.4, 157.6, 146.6, 143.9, 143.3, 142.9, 141.6, 138.2, 130.2, 129.8, 129.5, 127.9, 125.5, 125.1, 56.6, 50.9, 38.7, 37.2, 27.4, 26.5, 25.7, 25.2, 18.1, 14.2, 14.2, 11.7; HRMS (ESI) calcd. for $C_{30}H_{37}N_3O$ $[M + H]^+$: 456.3009. Found: 456.3023. The diastereomeric excess (>99% de) was determined by HPLC (XTerra MS C-18 column; gradient 40−60% of (0.1%TFA/MeCN)/(0.1%TFA/water); flow rate 1 mL/min; detection 254 nm; retention time 5.66 min).

*(1S,2S)-2-(Pyridin-2-yl)cyclopropanecarboxylic Acid [(2S,3S)-2-Amino-3-methylpentyl]-(4′-propylbiphenyl-4-yl)amide Dihydrochloride (**3**).* The procedure for **15a** was followed using 120 mg (0.23 mmol) of (1*S*,2*S*)-**14**, followed by deprotection of the Boc protecting group with 4 M HCl in dioxane, to give 98 mg (81% over two steps) of **3** as a white solid: mp 122 °C (fusion); $[\alpha]^{23}_{D}$ −91.3° (*c* 1, CH_3OH); ^1H NMR (300 MHz; CD_3OD) δ 8.62 (br d, *J* = 3.0 Hz, 1H), 8.31 (br t, *J* = 7.5 Hz, 1H), 7.80−7.58 (m, 6H), 7.49 (d, *J* = 9.0 Hz, 2H), 7.26 (d, *J* = 9.0 Hz, 2H), 4.46 (dd, *J* = 15.0, 9.0 Hz, 1H), 3.68 (d, *J* = 15.0 Hz, 1H), 3.46−3.38 (m, 1H), 3.12−3.02 (m, 1H), 2.61 (t, *J* = 7.5 Hz, 2H), 2.25−2.05 (m, 1H), 1.98−1.86 (m, 1H), 1.86−1.60 (m, 4H), 1.45−1.15 (m, 2H), 1.10−0.90 (m, 6H), 0.82 (t, *J* = 6.0 Hz, 3H); ^{13}C NMR (75 MHz; CD_3OD) δ 173.3, 157.5, 146.7, 143.9, 143.3, 142.9, 141.4, 138.2, 130.2, 130.0, 129.4, 127.9, 125.6, 125.3, 56.5, 50.6, 38.7, 37.4, 27.0, 26.7, 25.7, 25.6, 17.2, 14.1, 11.8; HRMS (ESI) calcd. for $C_{30}H_{37}N_3O$ $[M + H]^+$: 456.3009. Found: 456.3018. The diastereomeric excess (>99% de) was determined by HPLC (XTerra MS C-18 column; gradient 40−60% of (0.1%TFA/MeCN)/(0.1%TFA/water); flow rate 1 mL/min; detection 254 nm; retention time 6.35 min).

*tert-Butyl {2-[(4-Bromophenyl)amino]ethyl}carbamate (**18a**).* The procedure for **13** was followed using 1.03 g (5.96 mmol) of **11** and 0.95 g (5.96 mmol) of *N*-Boc-2-aminoacetaldehyde (**17a**) to give 1.38 g (50%) of **18a** as a yellow oily residue: ^1H NMR (300 MHz; $CDCl_3$) δ 7.23 (d, *J* = 9.0 Hz, 2H), 6.48 (d, *J* = 9.0 Hz, 2H), 4.80 (br s, 1H), 4.10 (br s, 1H), 3.41−3.30 (m, 2H), 3.30−3.26 (m, 2H), 1.45 (s, 9H); ^{13}C NMR (75 MHz; $CDCl_3$) δ 156.5, 147.1, 131.9, 114.2, 108.8, 79.6, 44.4, 40.0, 28.4; MS (ESI) *m*/*z* 315.1 $[M + H]^+$ (^{79}Br), 317.2 $[M + H]^+$ (^{81}Br).

*tert-Butyl {(2S)-1-[(4-Bromophenyl)amino]propan-2-yl}carbamate (**18b**).* The procedure for **13** was followed using 247 mg (1.44 mmol) of **11** and 250 mg (1.44 mmol) of *N*-Boc-L-alaninal (**17b**) to give 410 mg (86%) of **18b** as a white solid: mp 115−117 °C; $[\alpha]^{23}_{D}$ +3.1° (*c* 0.52, CH_3OH); ^1H NMR (300 MHz; $CDCl_3$) δ 7.23 (d, *J* = 9.0 Hz, 2H), 6.48 (d, *J* = 9.0 Hz, 2H), 4.48 (br s, 1H), 4.12 (br s, 1H), 4.00−3.86 (m, 1H), 3.18−2.98 (m, 2H), 1.45 (s, 9H), 1.21 (d, *J* = 6.0 Hz, 3H); ^{13}C NMR (75 MHz; $CDCl_3$) δ 156.0, 147.3, 131.9, 114.1, 108.7, 79.7, 50.6, 46.3, 28.4, 19.0; MS (ESI) *m*/*z* 329.3 $[M + H]^+$ (^{79}Br), 331.2 $[M + H]^+$ (^{81}Br).

*tert-Butyl {(2S)-1-[(4′-Propylbiphenyl-4-yl)amino]-3-methylbutan-2-yl}carbamate (**18c**).* The procedure for **13** was followed using 150 mg (0.71 mmol) of 4-(4′-propylphenyl)aniline (**16**) and 143 mg (0.71 mmol) of aldehyde **17c**, prepared by oxidation of *N*-Boc-L-valinol, to give 185 mg (66%) of **18c** as a white solid: mp 83−85 °C; $[\alpha]^{23}_{D}$ +22.2° (*c* 0.54, CH_3OH); ^1H NMR (300 MHz; $CDCl_3$) δ 7.45 (d, *J* = 9.0 Hz, 2H), 7.42 (d, *J* = 9.0 Hz, 2H), 7.20 (d, *J* = 9.0 Hz, 2H), 6.65 (d, *J* = 9.0 Hz, 2H), 4.51 (d, *J* = 6.0 Hz, 1H), 4.12 (br s, 1H), 3.80−3.65 (m, 1H), 3.35−3.20 (m, 1H), 3.12−3.00 (m, 1H), 2.60 (t, *J* = 7.5 Hz, 2H), 1.95−1.80 (m, 1H), 1.75−1.58 (m, 2H), 1.45 (s, 9H), 1.05−0.90 (m, 9H); ^{13}C NMR (75 MHz; $CDCl_3$) δ 156.8, 147.8, 140.5, 138.8, 130.3, 128.9, 127.8, 126.2, 113.0, 79.5, 55.7, 47.1, 37.8, 30.6, 28.5, 24.7, 19.6, 18.2, 14.0; MS (ESI) *m*/*z* 397.5 $[M + H]^+$.

*tert-Butyl {(2S)-1-[(4′-Propylbiphenyl-4-yl)amino]-4-methylpentan-2-yl}carbamate (**18d**).* The procedure for **13** was followed using 90 mg (0.43 mmol) of **16** and 93 mg (0.43 mmol) of aldehyde **17d**, prepared by oxidation of *N*-Boc-L-leucinol, to give 120 mg (68%) of **18d** as a white solid: mp 98−100 °C; $[\alpha]^{23}_{D}$ +2.4° (*c* 0.54, CH_3OH); ^1H NMR (300 MHz; $CDCl_3$) δ 7.45 (d, *J* = 9.0 Hz, 2H), 7.42 (d, *J* = 9.0 Hz, 2H), 7.22 (d, *J* = 9.0 Hz, 2H), 6.66 (d, *J* = 9.0 Hz, 2H), 4.41 (br s, 1H), 4.21 (br s, 1H), 4.00−3.82 (m, 1H), 3.32−3.20 (m, 1H), 3.12−3.00 (m, 1H), 2.62 (t, *J* = 7.5 Hz, 2H), 1.80−1.60 (m, 3H), 1.45 (s, 9H), 1.43−1.35 (m, 2H), 1.05−0.90 (m, 9H); ^{13}C NMR (75 MHz; $CDCl_3$) δ 156.3, 147.7, 140.5, 138.7, 130.2, 128.7, 127.8, 126.1, 112.9, 79.5, 49.9, 49.0, 42.5, 37.7, 28.4, 25.0, 24.6, 23.1, 22.1, 13.9; MS (ESI) *m*/*z* 411.5 $[M + H]^+$.

*tert-Butyl {(2S)-1-[(4-Bromophenyl)amino]-3-phenylpropan-2-yl}carbamate (**18e**).* The procedure for **13** was followed using 172 mg (1.00 mmol) of **11** and 250 mg (1.00 mmol) of *N*-Boc-L-phenylalaninal (**17e**) to give 350 mg (86%) of **18e** as a white solid: mp 124−126 °C; $[\alpha]^{23}_{D}$ +12.2° (*c* 0.53, CH_3OH); ^1H NMR (300 MHz; $CDCl_3$) δ 7.36−7.15 (m, 7H), 6.43 (d, *J* = 9.0 Hz, 2H), 4.51 (br s, 1H), 4.15−3.98 (m, 2H), 3.26−3.15 (m, 1H), 3.10−2.92 (m, 1H), 2.96−1.80 (m, 2H), 1.41 (s, 9H); ^{13}C NMR (75 MHz; $CDCl_3$) δ 156.0, 147.1, 137.3, 131.9, 129.2, 128.7, 126.8, 114.3, 109.0, 79.8, 51.4, 47.9, 39.2, 28.3; MS (ESI) *m*/*z* 405.3 $[M + H]^+$ (^{79}Br), 407.3 $[M + H]^+$ (^{81}Br).

tert-Butyl (2-{4-Bromophenyl-[(1R,2R*)-2-(pyridin-2-yl)-cyclopropanecarbonyl]amino}ethyl)carbamate (**19a**).* The procedure for **14** was followed using 180 mg (0.57 mmol) of **18a** and 136 mg (0.68 mmol) of racemic **10** to give 140 mg (53%) of **19a** as a yellow oil: ^1H NMR (300 MHz; $CDCl_3$) δ 8.31 (d, *J* = 6.0 Hz, 1H), 7.60−7.50 (m, 1H), 7.43 (d, *J* = 9.0 Hz, 2H), 7.21−7.00 (m, 4H), 5.05 (br s, 1H), 3.92−3.80 (m, 2H), 3.40−3.25 (m, 2H), 2.68−2.58 (m, 1H), 2.00−1.90 (m, 1H), 1.70−1.58 (m, 1H), 1.41 (s, 9H), 1.20−1.05 (m, 1H); MS (ESI) *m*/*z* 460.3 $[M + H]^+$ (^{79}Br), 462.1 $[M + H]^+$ (^{81}Br). This product contained impurity as judged by ^1H NMR analysis, which was used in the next step without further purification.

tert-Butyl [(2S)-1-{4-Bromophenyl-[(1R,2R*)-2-(pyridin-2-yl)-cyclopropanecarbonyl]amino}propan-2-yl]carbamate (**19b**).* The procedure for **14** was followed using 99 mg (0.30 mmol) of **18b** and 72 mg (0.36 mmol) of racemic **10** to give 85 mg (60%) of **19b** as a 1:1 diastereomeric mixture: ^1H NMR (300 MHz; $CDCl_3$) δ 8.30 (d, *J* = 6.0 Hz, 1H), 7.58−7.36 (m, 3H), 7.20−6.95 (m, 4H), 4.98 (d, *J* = 6.0 Hz, 1H), 4.22−4.05 (m, 1H), 3.92−3.70 (m, 1H), 3.32−3.18 (m, 2H), 2.68−2.60 (m, 0.5H), 2.60−2.46 (m, 0.5H), 1.95−1.80 (m, 1H), 1.68−1.52 (m, 1H), 1.43 and 1.41 (2s, 9H), 1.10 (d, *J* = 6.0 Hz, 3H); MS (ESI) *m*/*z* 474.4 $[M + H]^+$ (^{79}Br), 476.5 $[M + H]^+$ (^{81}Br).

tert-Butyl (2S)-1-{(4′-Propylbiphenyl-4-yl)-[(1R,2R*)-2-(pyridin-2-yl)cyclopropanecarbonyl]amino}-3-methylbutan-2-yl}carbamate (**19c**).* The procedure for **14** was followed using 120 mg (0.34 mmol) of **18c** and 81 mg (0.40 mmol) of racemic **10** to give 110 mg (60%) of **19c** as a 1:1 diastereomeric mixture: ^1H NMR (300 MHz; $CDCl_3$) δ 8.26−8.18 (m, 1H), 7.55−7.34 (m, 5H), 7.30−7.21 (m, 5H), 6.98−6.90 (m, 1H), 5.08−4.92 (m, 1H), 4.45−4.32 (m, 1H), 3.80−3.60 (m, 1H), 3.25−3.12 (m, 1H), 2.74−2.48 (m, 3H), 2.08−1.92 (m, 1H),

dx.doi.org/10.1021/cn500082p | ACS Chem. Neurosci. 2014, 5, 576−587

1.78−1.52 (m, 5H), 1.47 and 1.43 (2s, 9H), 1.00−0.80 (m, 9H); MS (ESI) m/z 542.6 $[M + H]^+$.

tert-Butyl (2S)-1-{(4′-Propylbiphenyl-4-yl)-[(1R,2R*)-2-(pyridin-2-yl)cyclopropanecarbonyl]amino}-4-methylpentan-2-yl}-carbamate (**19d**).* The procedure for **14** was followed using 90 mg (0.22 mmol) of **18d** and 53 mg (0.26 mmol) of racemic **10** to give 65 mg (53%) of **19d** as a 1:1 diastereomeric mixture: ^{1}H NMR (300 MHz; $CDCl_3$) δ 8.28−8.20 (m, 1H), 7.55−7.35 (m, 5H), 7.32−7.12 (m, 5H), 7.00−6.90 (m, 1H), 4.90−4.80 (m, 1H), 4.28−4.12 (m, 1H), 4.00−3.80 (m, 1H), 3.38−3.20 (m, 1H), 2.75−2.50 (m, 3H), 2.10−1.90 (m, 1H), 1.76−1.52 (m, 5H), 1.45 and 1.41 (2s, 9H), 1.40−1.12 (m, 2H), 1.02−0.80 (m, 9H); MS (ESI) m/z 556.8 $[M + H]^+$.

tert-Butyl [(2S)-1-{4-Bromophenyl-[(1R,2R*)-2-(pyridin-2-yl)-cyclopropanecarbonyl]amino}-3-phenylpropan-2-yl]carbamate (**19e**).* The procedure for **14** was followed using 99 mg (0.30 mmol) of **18e** and 72 mg (0.36 mmol) of racemic **10** to give 85 mg (60%) of **19e** as a 1:1 diastereomeric mixture: ^{1}H NMR (300 MHz; $CDCl_3$) δ 8.29 (d, J = 6.0 Hz, 1H), 7.55−7.30 (m, 3H), 7.30−6.90 (m, 9H), 5.05−4.92 (m, 1H), 4.28−3.90 (m, 2H), 3.30−3.20 (m, 1H), 2.88−2.48 (m, 3H), 1.95−1.82 (m, 1H), 1.70−1.50 (m, 1H), 1.39 and 1.37 (2s, 9H), 1.32−1.20 (m, 1H); MS (ESI) m/z 550.4 $[M + H]^+$ (^{79}Br), 552.5 $[M + H]^+$ (^{81}Br).

tert-Butyl (2-{(4′-Propylbiphenyl-4-yl)-[(1R,2R*)-2-(pyridin-2-yl)-cyclopropanecarbonyl]amino}ethyl)carbamate (**20a**).* The procedure for **15a** was followed using 140 mg (0.30 mmol) of **19a** and 83 mg (0.47 mmol) of 4-propylphenylboronic acid to give 80 mg (53%) of **20a**: ^{1}H NMR (300 MHz; $CDCl_3$) δ 8.22 (d, J = 6.0 Hz, 1H), 7.55−7.40 (m, 5H), 7.25−7.12 (m, 5H), 7.00−6.90 (m, 1H), 5.17 (br s, 1H), 3.90 (t, J = 6.0 Hz, 2H), 3.42−3.25 (m, 3H), 2.62 (t, J = 7.5 Hz, 2H), 2.10−1.98 (m, 1H), 1.75−1.60 (m, 2H), 1.55−1.35 (m, 2H), 1.42 (s, 9H), 0.97 (t, J = 6.0 Hz, 3H); ^{13}C NMR (75 MHz; $CDCl_3$) δ 172.9, 159.4, 156.1, 149.3, 142.3, 141.0, 137.4, 135.8, 129.7, 129.0, 128.2, 128.1, 126.9, 122.4, 121.0, 79.1, 49.2, 39.7, 37.7, 28.4, 27.7, 24.9, 24.5, 17.7, 13.9; MS (ESI) m/z 500.8 $[M + H]^+$.

tert-Butyl [(2S)-1-{(4′-Propylbiphenyl-4-yl)-[(1R,2R*)-2-(pyridin-2-yl)cyclopropanecarbonyl]amino}propan-2-yl]carbamate (**20b**).* The procedure for **15a** was followed using 50 mg (0.11 mmol) of **19b** and 29 mg (0.16 mmol) of 4-propylphenylboronic acid to give 45 mg (80%) of **20b** as a 1:1 diastereomeric mixture: ^{1}H NMR (300 MHz; $CDCl_3$) δ 8.28−8.18 (m, 1H), 7.56−7.38 (m, 5H), 7.28−7.10 (m, 5H), 7.00−6.90 (m, 1H), 5.20−5.06 (m, 1H), 4.32−4.15 (m, 1H), 3.96−3.78 (m, 1H), 3.38−3.22 (m, 1H), 2.70−2.48 (m, 3H), 2.06−1.90 (m, 2H), 1.78−1.58 (m, 3H), 1.45 and 1.43 (2s, 9H), 1.12 (d, J = 6.0 Hz, 3H), 0.97 (t, J = 7.5 Hz, 3H); MS (ESI) m/z 514.6 $[M + H]^+$.

tert-Butyl [(2S)-1-{(4′-Propylbiphenyl-4-yl)-[(1R,2R*)-2-(pyridin-2-yl)cyclopropanecarbonyl]amino}-3-phenylpropan-2-yl]-carbamate (**20e**).* The procedure for **15a** was followed using 90 mg (0.16 mmol) of **19e** and 44 mg (0.24 mmol) of 4-propylphenylboronic acid to give 75 mg (80%) of **20e** as a 1:1 diastereomeric mixture: ^{1}H NMR (300 MHz; $CDCl_3$) δ 8.26−8.15 (m, 1H), 7.55−7.38 (m, 6H), 7.28−7.05 (m, 9H), 7.00−6.90 (m, 1H), 5.16−5.05 (m, 1H), 4.35−4.18 (m, 1H), 4.15−3.95 (m, 1H), 3.34−3.22 (m, 1H), 2.88−2.45 (m, 5H), 2.05−1.90 (m, 1H), 1.75−1.50 (m, 3H), 1.41 and 1.40 (2s, 9H), 1.30−1.20 (m, 1H), 0.97 (t, J = 7.5 Hz, 3H); MS (ESI) m/z 590.8 $[M + H]^+$.

(1R,2R*)-2-(Pyridin-2-yl)cyclopropanecarboxylic Acid [2-Amino-ethyl-(4′-propylbiphenyl-4-yl)amide Dihydrochloride (**5a**).* The procedure for **1** was followed using 60 mg (0.12 mmol) of **20a** and 1 mL of 4 M HCl in dioxane to give 52 mg (92%) of **5a** as a light yellow foam: ^{1}H NMR (300 MHz; CD_3OD) δ 8.56 (br s, 1H), 8.23 (br t, 1H), 7.50−7.63 (m, 3H), 7.62−7.43 (m, 5H), 7.27 (d, J = 9.0 Hz, 2H), 4.25−3.98 (m, 2H), 3.22−3.10 (m, 2H), 3.00−2.78 (m, 1H), 2.63 (t, J = 7.5 Hz, 2H), 2.18−2.08 (m, 1H), 1.94−182 (m, 1H), 1.72−1.60 (m, 3H), 0.96 (t, J = 7.5 Hz, 3H); ^{13}C NMR (75 MHz; CD_3OD) δ 173.1, 157.9, 146.2, 144.1, 143.9, 143.0, 141.4, 138.2, 130.2, 130.0, 129.6, 128.0, 125.5, 125.3, 39.9, 38.7, 27.1, 25.8, 25.7, 18.0, 14.1; HRMS (ESI) calcd. for $C_{26}H_{29}N_3O$ $[M + H]^+$: 400.2383. Found: 400.2388.

(1R,2R*)-2-(Pyridin-2-yl)cyclopropanecarboxylic Acid [(2S)-2-Aminopropyl-(4′-propylbiphenyl-4-yl)amide Dihydrochloride (**5b**).* The procedure for **1** was followed using 40 mg (0.078 mmol) of **20b** and 1 mL of 4 M HCl in dioxane to give 37 mg (98%) of **5b** as a 1:1 diastereomeric mixture: ^{1}H NMR (300 MHz; CD_3OD) δ 8.70−8.58 (m, 1H), 8.40−8.28 (m, 1H), 7.85−7.40 (m, 8H), 7.30−7.20 (m, 2H), 4.42−4.26 (m, 0.5H), 4.25−4.10 (m, 0.5H), 3.95−3.82 (m, 0.5H), 3.78−3.48 (m, 1.5H), 3.10−2.90 (m, 1H), 2.62 (t, J = 7.5 Hz, 2H), 2.22−2.06 (m, 1H), 2.05−1.85 (m, 1H), 1.78−1.68 (m, 3H), 1.42−1.20 (m, 3H), 0.95 (t, J = 6.0 Hz, 3H); HRMS (ESI) calcd. for $C_{27}H_{31}N_3O$ $[M + H]^+$: 414.2540. Found: 414.2547.

(1R,2R*)-2-(Pyridin-2-yl)cyclopropanecarboxylic Acid [(2S)-2-Amino-3-methylbutyl]-(4′-propylbiphenyl-4-yl)amide Dihydrochloride (**5c**).* The procedure for **1** was followed using 60 mg (0.11 mmol) of **19c** and 1 mL of 4 M HCl in dioxane to give 54 mg (95%) of **5c** as a 1:1 diastereomeric mixture: ^{1}H NMR (300 MHz; CD_3OD) δ 8.55−8.38 (m, 1H), 8.22−8.10 (m, 1H), 7.70−7.30 (m, 8H), 7.25−7.10 (m, 2H), 4.38−4.25 (m, 0.5H), 4.24−4.10 (m, 0.5H), 3.88−3.40 (m, 2H), 3.30−3.10 (m, 1H), 2.98−2.80 (m, 1H), 2.52 (t, J = 7.5 Hz, 2H), 2.16−1.78 (m, 3H), 1.68−1.46 (m, 2H), 1.00−0.75 (m, 9H); HRMS (ESI) calcd. for $C_{29}H_{35}N_3O$ $[M + H]^+$: 442.2853. Found: 442.2856.

(1R,2R*)-2-(Pyridin-2-yl)cyclopropanecarboxylic Acid [(2S)-2-Amino-4-methylpentyl]-(4′-propylbiphenyl-4-yl)amide Dihydrochloride (**5d**).* The procedure for **1** was followed using 50 mg (0.09 mmol) of **19d** and 1 mL of 4 M HCl in dioxane to give 46 mg (97%) of **5d** as a 1:1 diastereomeric mixture: ^{1}H NMR (300 MHz; CD_3OD) δ 8.60−8.50 (m, 1H), 8.30−8.18 (m, 1H), 7.78−7.40 (m, 8H), 7.27 (d, J = 9.0 Hz, 2H), 4.36−4.20 (m, 0.5H), 4.20−4.18 (m, 0.5H), 4.05−3.90 (m, 0.5H), 3.88−3.60 (m, 1.5H), 3.52−3.40 (m, 1H), 3.10−2.90 (m, 1H), 2.62 (t, J = 7.5 Hz, 2H), 2.22−2.08 (m, 1H), 2.00−1.88 (m, 1H), 1.76−1.42 (m, 5H), 1.02−0.75 (m, 9H); HRMS (ESI) calcd. for $C_{30}H_{37}N_3O$ $[M + H]^+$: 456.3009. Found: 456.3017.

(1R,2R*)-2-(Pyridin-2-yl)cyclopropanecarboxylic Acid [(2S)-2-Amino-3-phenylpropyl]-(4′-propylbiphenyl-4-yl)amide Dihydrochloride (**5e**).* The procedure for **1** was followed using 70 mg (0.12 mmol) of **20e** and 1 mL of 4 M HCl in dioxane to give 65 mg (96%) of **5e** as a 1:1 diastereomeric mixture: ^{1}H NMR (300 MHz; CD_3OD) δ 8.65−8.58 (m, 1H), 8.40−8.26 (m, 1H), 7.82−7.70 (m, 1H), 7.70−7.36 (m, 7H), 7.35−7.05 (m, 7H), 4.45−4.30 (m, 0.5H), 4.25−4.10 (m, 0.5H), 4.00−3.86 (m, 0.5H), 3.85−3.55 (m, 1.5H), 3.16−2.88 (m, 3H), 2.69 (t, J = 7.5 Hz, 2H), 2.22−2.05 (m, 1H), 2.05−1.88 (m, 1H), 1.76−1.58 (m, 3H), 0.95 (t, J = 7.5 Hz, 3H); HRMS (ESI) calcd. for $C_{33}H_{35}N_3O$ $[M + H]^+$: 490.2853. Found: 490.2859.

*tert-Butyl [(2S,3S)-1-{4-Bromophenyl-[(1R,2R)-2-phenyl-1-cyclopropanecarbonyl]amino}-3-methylpentan-2-yl]carbmate (**21a**).* A solution of (1*R*,2*R*)-2-phenyl-1-cyclopropanecarboxylic acid[12] (97 mg, 0.6 mmol) in thionyl chloride (3 mL) was refluxed overnight. After cooling to room temperature, the mixture was concentrated under reduced pressure. The resulting residue was dissolved in CH_2Cl_2 (5 mL) and treated with **13** (150 mg, 0.4 mmol) and Et_3N (0.17 mL, 1.2 mmol). The reaction mixture was stirred at room temperature overnight. Saturated $NaHCO_3$ (5 mL) was added and the layers were separated. The aqueous layer was extracted with CH_2Cl_2 (3 × 10 mL). The combined organic layers were washed with brine (3 × 10 mL), dried (Na_2SO_4), and concentrated under reduced pressure. Flash column chromatography of the crude product on silica gel using 0−25% EtOAc in hexanes afforded **21a** (155 mg, 75%) as a white foam: $[\alpha]^{23}_D$ +35.2° (c 0.52, CH_3OH); ^{1}H NMR (300 MHz; $CDCl_3$) δ 7.49 (d, J = 9.0 Hz, 2H), 7.25−7.10 (m, 5H), 6.94 (d, J = 9.0 Hz, 2H), 4.96 (d, J = 12.0 Hz, 1H), 4.37 (dd, J = 12.0, 10.5 Hz, 1H), 3.80−3.66 (m, 1H), 3.13 (dd, J = 12.0, 3.0 Hz, 1H), 2.66−2.55 (m, 1H), 1.61−1.48 (m, 4H), 1.45 (s, 9H), 1.16−1.10 (m, 2H), 0.91−0.82 (m, 6H); ^{13}C NMR (75 MHz; $CDCl_3$) δ 173.3, 156.4, 141.4, 140.4, 133.1, 130.2, 128.5, 126.4, 122.0, 79.0, 54.1, 50.3, 38.3, 28.6, 26.4, 25.4, 24.1, 17.8, 15.3, 11.9; MS (ESI) m/z 515.4 $[M + H]^+$ (^{79}Br), 517.4 $[M + H]^+$ (^{81}Br).

*tert-Butyl [(2S,3S)-1-{4-Bromophenyl-[(pyridin-2-yl)-methylcarbonyl]amino}-3-methylpentan-2-yl]carbamate (**21b**).* To a solution of **13** (200 mg, 0.54 mmol) in CH_3CN (10 mL) at room temperature were added 2-pyridylacetic acid hydrochloride (113 mg, 0.65 mmol), Et_3N (0.23 mL, 1.6 mmol), and HBTU (246 mg, 0.65 mmol). The reaction mixture was stirred at room temperature overnight. The solution was diluted with EtOAc (50 mL) and washed with saturated $NaHCO_3$ (10 mL) and brine (10 mL). The organic

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phase was dried (Na_2SO_4) and concentrated under reduced pressure. Flash column chromatography of the crude product on silica gel using 0−25% EtOAc in hexanes afforded **21b** (170 mg, 64%) as a brown residue: ^{1}H NMR (300 MHz; $CDCl_3$) δ 8.40 (d, J = 3.0 Hz, 1H), 7.56−7.48 (m, 1H), 7.47 (d, J = 9.0 Hz, 2H), 7.13−6.98 (m, 4H), 4.89 (d, J = 9.0 Hz, 1H), 4.30 (t, J = 12.0 Hz, 1H), 3.70−3.55 (m, 1H), 3.54 (s, 2H), 3.06 (dd, J = 15.0, 3.0 Hz, 1H), 1.50−1.42 (m, 2H), 1.41 (s, 9H), 1.10−0.92 (m, 1H), 0.83−0.70 (m, 6H); ^{13}C NMR (75 MHz; $CDCl_3$) δ 169.9, 155.1, 154.5, 148.3, 140.1, 135.3, 132.0, 129.2, 122.8, 121.3, 120.7, 77.9, 52.5, 49.0, 42.9, 37.0, 27.4, 24.2, 14.0, 10.7; MS (ESI) m/z 490.8 $[M + H]^+$ (^{79}Br), 492.6 $[M + H]^+$ (^{81}Br).

*tert-Butyl [(2S,3S)-1-{(4′-Propylbiphenyl-4-yl)-[(1R,2R)-2-phenyl-1-cyclopropanecarbonyl]amino}-3-methylpentan-2-yl]carbamate (**22a**).* The procedure for **15a** was followed using 100 mg (0.19 mmol) of **21a** and 54 mg (0.32 mmol) of 4-propylphenylboronic acid to give 80 mg (76%) of **22a** as a white foam: $[\alpha]^{23}_D$ +71.4° (c 0.52, CH_3OH); ^{1}H NMR (300 MHz; $CDCl_3$) δ 7.56 (d, J = 9.0 Hz, 2H), 7.48 (d, J = 9.0 Hz, 2H), 7.36−7.22 (m, 4H), 7.21−7.01 (m, 3H), 6.94 (d, J = 9.0 Hz, 2H), 5.08 (d, J = 12.0 Hz, 1H), 4.43 (t, J = 12.0 Hz, 1H), 3.86−3.72 (m, 1H), 3.19 (dd, J = 13.5, 4.5 Hz, 1H), 2.70−2.56 (m, 3H), 1.76−1.55 (m, 6H), 1.47 (s, 9H), 1.18−1.04 (m, 2H), 1.00 (t, J = 7.5 Hz, 3H), 0.93−0.82 (m, 6H); ^{13}C NMR (75 MHz; $CDCl_3$) δ 173.5, 156.4, 142.3, 141.0, 140.7, 140.5, 137.3, 129.0, 128.6, 128.3, 128.1, 126.9, 126.4, 126.2, 78.7, 54.1, 50.0, 38.2, 37.7, 28.5, 26.1, 25.3, 24.5, 23.9, 17.6, 15.1, 13.8, 11.8; MS (ESI) m/z 555.8 $[M + H]^+$.

*tert-Butyl [(2S,3S)-1-{(4′-Propylbiphenyl-4-yl)[(pyridin-2-yl)-methylcarbonyl]amino}-3-methylpentan-2-yl]carbamate (**22b**).* The procedure for **15a** was followed using 170 mg (0.35 mmol) of **21b** and 96 mg (0.54 mmol) of 4-propylphenylboronic acid to give 101 mg (55%) of **22b** as an oil: ^{1}H NMR (300 MHz; $CDCl_3$) δ 8.47 (d, J = 3.0 Hz, 1H), 765−7.56 (m, 3H), 7.50 (d, J = 9.0 Hz, 2H), 7.42−7.08 (m, 6H), 5.08 (d, J = 9.0 Hz, 1H), 4.44 (t, J = 12.0 Hz, 1H), 3.86−3.74 (m, 1H), 3.69 (s, 2H), 3.08 (dd, J = 15.0, 3.0 Hz, 1H), 2.63 (t, J = 7.5 Hz, 2H), 1.76−1.65 (m, 2H), 1.64−1.47 (m, 2H), 1.45 (s, 9H), 1.16−1.02 (m, 1H), 0.97 (t, J = 7.5 Hz, 3H), 0.92−0.78 (m, 6H); ^{13}C NMR (75 MHz; $CDCl_3$) δ 171.3, 156.2, 155.9, 149.2, 142.4, 141.2, 140.9, 137.3, 130.2, 129.0, 128.7, 128.3, 126.9, 123.9, 121.6, 78.8, 53.7, 49.9, 43.9, 38.0, 37.7, 28.5, 25.3, 24.5, 15.0, 13.8, 11.8; MS (ESI) m/z 530.9 $[M + H]^+$.

*(1R,2R)-2-Phenyl-1-cyclopropanecarboxylic Acid [(2S,3S)-2-Amino-3-methylpentyl]-(4′-propylbiphenyl-4-yl)amide Hydrochloride (**6a**).* The procedure for **1** was followed using 60 mg (0.11 mmol) of **22a** and 1 mL of 4 M HCl in dioxane to give 52 mg (96%) of **6a** as a white solid: mp 112 °C (fusion); $[\alpha]^{23}_D$ −5.7° (c 0.53, CH_3OH); ^{1}H NMR (300 MHz; CD_3OD) δ 7.65 (br d, J = 6.0 Hz, 2H), 7.56−7.38 (m, 4H), 7.26 (d, J = 9.0 Hz, 2H), 7.20−7.06 (m, 3H), 6.93 (d, J = 9.0 Hz, 2H), 4.38−4.22 (m, 1H), 3.83−3.60 (m, 1H), 3.44−3.33 (m, 1H), 2.64 (d, J = 7.5 Hz, 2H), 2.54−2.42 (m, 1H), 1.85−1.60 (m, 4H), 1.50−1.16 (m, 4H), 1.03−0.93 (m, 6H), 0.87 (t, J = 6.0 Hz, 3H); ^{13}C NMR (75 MHz; CD_3OD) δ 176.0, 143.8, 142.7, 142.0, 141.5, 138.4, 130.3, 129.8, 129.6, 129.5, 128.0, 127.5, 127.2, 57.0, 50.7, 38.7, 37.3, 28.1, 26.5, 26.0, 25.7, 17.9, 14.4, 14.2, 11.9; HRMS (ESI) calcd. for $C_{31}H_{38}N_2O$ $[M + H]^+$: 455.3057. Found: 455.3061.

*2-Pyridylacetic Acid [(2S,3S)-2-Amino-3-methylpentyl]-(4′-propylbiphenyl-4-yl)amide Dihydrochloride (**6b**).* The procedure for **1** was followed using 90 mg (0.17 mmol) of **22b** and 2 mL of 4 M HCl in dioxane to give 80 mg (94%) of **6b** as a white foam: ^{1}H NMR (300 MHz; CD_3OD) δ 8.76 (br d, J = 6.0 Hz, 1H), 8.41 (br t, J = 7.5 Hz, 1H), 7.92−7.50 (m, 8H), 7.30 (d, J = 6.0 Hz, 2H), 4.50−4.38 (m, 1H), 3.75−3.56 (m, 1H), 3.52−3.38 (m, 1H), 2.64 (d, J = 7.5 Hz, 2H), 1.90−1.60 (m, 3H), 1.50−1.15 (m, 2H), 1.02−0.90 (m, 6H), 0.84 (t, J = 6.0 Hz, 3H) (Methylene protons overlapped with methanol solvent peak); ^{13}C NMR (75 MHz; $CDCl_3$) δ 169.6, 150.2, 146.5, 142.7, 142.2, 141.6, 139.2, 136.8, 129.6, 129.1, 128.9, 126.9, 125.4, 55.2, 48.9, 39.8, 37.7, 35.8, 26.2, 24.4, 14.2, 13.8, 11.4; HRMS (ESI) calcd. for $C_{28}H_{35}N_3O$ $[M + H]^+$: 430.2853. Found: 430.2860.

Pharmacology. *Materials.* Isoproterenol was purchased from Sigma-Aldrich, and cell culture reagents (media, supplements, antibiotics, etc.) were purchased from Fisher Scientific. Human GPR88 cDNA was purchased from Missouri S&T cDNA Resource Center. The pGloSensor-22F plasmid was purchased from Promega.

Transient Transfection and cAMP Assay. HEK293T cells were transfected with human GPR88 cDNA and pGloSensor-22F overnight and plated in the poly-L-lysine coated 384-well white clear bottom cell culture plates using DMEM supplemented with 1% dialyzed fetal bovine serum at a density of 15,000 cells in 40 μL medium per well. The cell plates were incubated for 6−20 h before being used for assays. To measure receptor mediated Gα_i-activation, culture medium was removed and assay buffer (20 μL per well of 20 mM HEPES, 1× HBSS, pH 7.4) was added, followed by addition of 10 μL of test compound solution at serially diluted concentrations for 15 min at room temperature. After addition of 15 μL of Luciferin (4 mM final) and isoproterenol (200 nM final) and incubation for another 15 min, luminescence was read on the TriLux microbeta counter (PerkinElmer). To measure receptor mediated Gα_s-activation, cells were first incubated with test compound for 15 min followed by 15 μL of 4 mM Luciferin for another 15 min, and then luminescence counted as above.

GloSensor cAMP Assay Using Stable GPR88-22F HEK293 Cells. A GPR88-22F stable cell line was created by overexpressing the human GPR88 receptor and the pGloSensor-22F biosensor in HEK293 cells. The day before the assay, GPR88-22F cells were plated into 96-well white-walled assay plates at a density of 40 000 cells per well in culture medium (DMEM-HG supplemented with 10% FBS, 15 mM HEPES, and 100 units of penicillin/streptomycin). The plated cells were incubated overnight at 37 °C, 5% CO_2. The next day, the culture medium was gently removed and 100 μL of equilibration medium was gently added per the manufacturer's instructions (88% CO_2-independent medium, 10% FBS, 2% GloSensor cAMP reagent). The cells were incubated in the equilibration medium for 2 h at room temperature in the dark. Test compound dilutions were prepared at 11× concentration in 1× PBS and 10 μL was added to each appropriate well. Following 10 min at room temperature in the dark, 10 μL of 100 nM (final) isoproterenol (prepared at 12× concentration in 1× PBS) was added to each appropriate well. Following 30 min at room temperature in the dark, luminescence was read on the FlexStation III (1000 ms integration time, Molecular Devices).

Data Analysis. Relative luminescence units (RLU) were recorded and plotted against compound concentration. Data were fit to a three-parameter logistic function to generate EC_{50} values using GraphPad Prism software (San Diego, CA).

■ ASSOCIATED CONTENT

Ⓢ Supporting Information

Copies of HPLC results of **1**−**3**, **4a**−**i**, **5a**−**e**, **6a**, and **6b**. This material is available free of charge via the Internet at http://pubs.acs.org.

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Funding

We thank the National Institute of Mental Health (NIMH) for supporting the research at Psychoactive Drug Screening Program (PDSP). XPZ is grateful for financial support by NSF (CHE-1152767) and NIH (R01-GM098777).

Notes

The authors declare no competing financial interest.

■ ACKNOWLEDGMENTS

We thank Dr. Danni Harris for valuable discussions during the course of this work.

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