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Design, synthesis, and pharmacological evaluation of JDTic analogs to examine the significance of replacement of the 3-hydroxyphenyl group with pyridine or thiophene bioisosteres

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Abstract

The potent and selective KOR antagonist JDTic was derived from the *N*-substituted trans-3,4-dimethyl-4-(3-hydroxyphenyl)piperidine class of pure opioid antagonists. In previous studies we reported that compounds that did not have a hydroxyl on the 3-hydroxyphenyl group and did not have methyl groups at the 3- and 4-position of the piperidine ring were still potent and selective KOR antagonists. In this study we report JDTic analogs **2**, **3a–b**, **4a–b**, and **5**, where the 3-hydroxyphenyl ring has been replaced by a 2-, 3-, or 4-pyridyl or 3-thienyl group and do not have the 3-methyl or 3,4-dimethyl groups, remain potent and selective KOR antagonists. Of these, (3*R*)-7-hydroxy-*N*-(1*S*)-2-methyl-[4-methyl-4-pyridine-3-yl-carboxamide (**3b**) had the best overall binding potency and selectivity in a [^{35}S] GTPγS functional assay, with a K_e = 0.18 nM at the KOR and 273- and 16,700-fold selectivity for the KOR relative to the MOR and DOR, respectively. Calculated physiochemical properties for **3b** suggest that it will cross the blood-brain barrier.

Keywords: JDTic; Opioids; Kappa antagonist; ADME properties

1. Introduction

During the last 15-20 years considerable effort has been devoted by academic institutions, research institutions, and pharmaceutical companies to the development of potent and selective kappa opioid receptor (KOR) antagonists (see ref 1 for a review).[1] Animal behavioral studies have suggested that potent and selective KOR antagonists have potential as pharmacotherapies for treating mood disorders such as depression, anxiety, and substance abuse (nicotine, alcohol, cocaine, and opiates).

Abbreviations: [^{35}S]GTPγS, sulfur-35 guanosine-5′-*O*-(3-thio)triphosphate; DAMGO, [D-Ala^2,MePhe^4,Gly-ol^5]enkephalin; DPDPE, [D-Pen^2,D-Pen^5]enkephalin; U69,593, (5α,7α,8β)-(-)-*N*-methyl-*N*-[7-(1-pyrrolidinyl)-1-oxaspiro[4,5]dec-8-yl]benzeneacetamide

In our early studies we showed that (3*R*)-1,2,3,4-tetrahydro-7-hydroxy-*N*-[(1*S*)-1-[[(3*R*,4*R*)-4-(3-hydroxyphenyl)-3,4-dimethyl-1-piperidinyl]methyl]-2-methylpropyl]-3-isoquinolinecarboxamide (JDTic) was a potent and selective KOR antagonist.[2, 3] In more recent studies we reported that JDTic analog **1** (Figure 1), where both the 3- and 4-methyl groups were removed from JDTic, was still a potent and selective KOR antagonist.[4] In addition we reported that JDTic analogs with the hydroxyl group removed from the 4-(3-hydroxyphenyl) moiety were still potent and selective KOR antagonists.[5] The fact that **1** is a potent and selective KOR antagonist suggest that replacement of the 4-phenyl group in **1** with heterocyclic group could also lead to potent and selective KOR antagonist. In this study we report the synthesis, [^{35}S]GTPγS *in vitro* binding and calculated physiochemical properties of compounds **2**, **3a–b**, and **4a–b** which have a 2-, 3-, or 4-pyridyl group or **5** which has a thiophene ring replacing the phenyl ring in previously reported JDTic analogs (Figure 1).

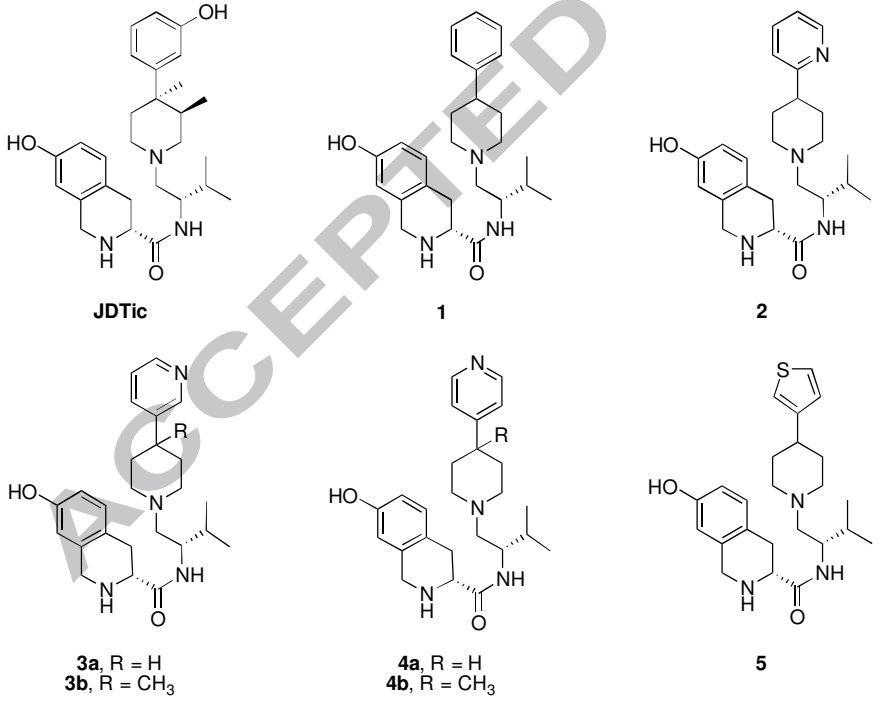
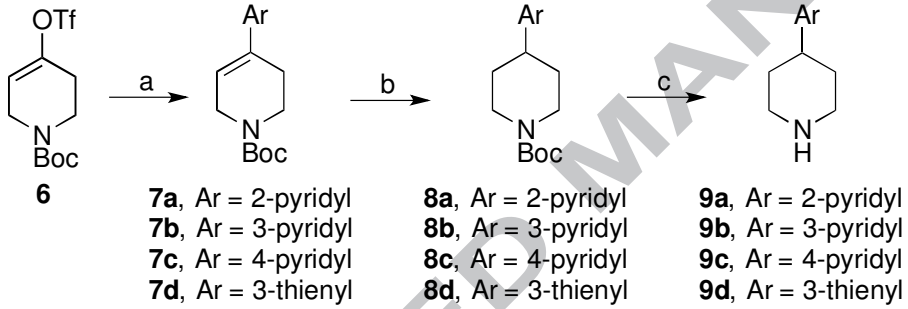


Figure 1. Structures of **JDTic**, **1**, **2**, **3a-b**, **4a-b**, and **5**.

2. Chemistry

The known piperidines **9a**,[6] **9b**,[7] **9c**,[8] and **9d** were prepared according to the route shown in Scheme 1, affording material consistent with literature characterization. Beginning with vinyl triflate **6**, Negishi or Suzuki cross-coupling afforded the pyridyl and thienyl tetrahydropyridine derivatives **7a–d**. Catalytic hydrogenation yielded the saturated *N*-Boc-piperidines **8a–d**. Treatment of **8a–d** with hydrogen chloride in dioxane deprotected the Boc to afford the amine hydrochloride salts **9a–d**, which were used directly in the next step.

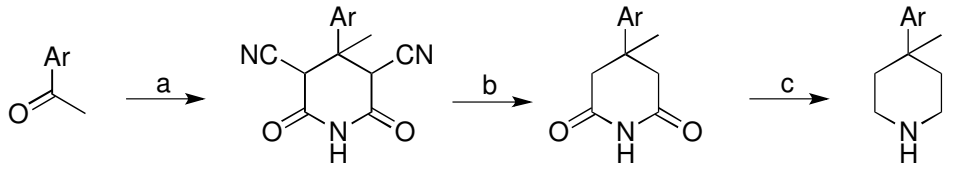
Scheme 1[a]



[a] Reagents: a) Suzuki or Negishi coupling; b) H_2, Pd/C; c) 4M HCl in dioxane, CH_3CN.

Amines **9e–f** were prepared according to the route shown in Scheme 2. Knoevenagel condensation of the acetylpyridines **10a–b** with ethyl cyanoacetate using ammonium acetate and acetic acid followed by condensation with 2-cyanoacetamide gave the intermediates **11a–b**. Hydrolysis and decarboxylation of **11a–b** using sulfuric acid gave glutaric acids which were not isolated. Upon heating the crude diacids neat with urea as a melt, the glutarimides **12a–b** were produced. Reduction of the imides with diborane in tetrahydrofuran afforded piperidines **9e–f**.

Scheme 2[a]



10a, Ar = 3-pyridyl
10b, Ar = 4-pyridyl

11a, Ar = 3-pyridyl
11b, Ar = 4-pyridyl

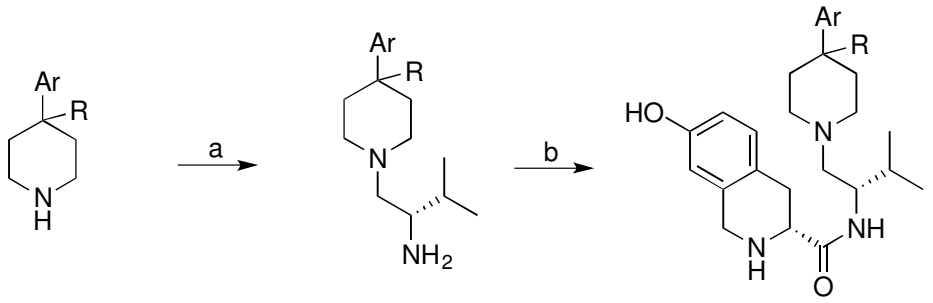
12a, Ar = 3-pyridyl
12b, Ar = 4-pyridyl

9e, Ar = 3-pyridyl
9f, Ar = 4-pyridyl

[a] Reagents: a) (1) Ethyl cyanoacetate, NH_4OAc, HOAc, (2) NaOEt, 2-cyanoacetamide; b) (1) H_2SO_4, (2) urea; c) $BH_3 \bullet S(CH_3)_2$, THF.

Coupling of piperidines **9a–f** with *N*-Boc-L-valine using 3-(3-dimethylaminopropyl)carbodiimide hydrochloride (EDC•HCl) with catalytic *N*-hydroxybenzotriazole (HOBt) or 2-(1H-benzotriazol-1-yl)-1,1,3,3-tetramethyluronium hexafluorophosphate (HBTU) in acetonitrile with triethylamine afforded the desired Boc-protected amides, which were deprotected with 4M hydrogen chloride in dioxane or hydrochloric acid in aqueous methanol and were directly reduced with diborane or borane dimethylsulfide in tetrahydrofuran to yield the amines **13a–f** (Scheme 3). The amines **13a–f** were coupled with (3*R*)-2-(*tert*-butoxycarbonyl)-7-hydroxy-1,2,3,4-tetrahydroisoquinoline-3-carboxylic (7-hydroxy-Boc-D-Tic-OH) using EDC•HCl with a catalytic amount of HOBt in dichloromethane to give the Boc-protected intermediates. The desired final products **2**, **3a–b**, **4a–b**, and **5** resulted from treatment of an acetonitrile solution of the Boc-intermediates with 4M hydrogen chloride in dioxane.

Scheme 3[a]



9a, Ar = 2-pyridyl, R = H	**13a**, Ar = 2-pyridyl, R = H	**2**, Ar = 2-pyridyl, R = H
9b, Ar = 3-pyridyl, R = H	**13b**, Ar = 3-pyridyl, R = H	**3a**, Ar = 3-pyridyl, R = H
9c, Ar = 4-pyridyl, R = H	**13c**, Ar = 4-pyridyl, R = H	**4a,** Ar = 4-pyridyl, R = H
9d, Ar = 3-thienyl, R = H	**13d**, Ar = 3-thienyl, R = H	**5**, Ar = 3-thienyl, R = H
9e, Ar = 3-pyridyl, R = CH_3	**13e**, Ar = 3-pyridyl, R = CH_3	**3b**, Ar = 3-pyridyl, R = CH_3
9f, Ar = 4-pyridyl, R = CH_3	**13f**, Ar = 4-pyridyl, R = CH_3	**4b**, Ar = 4-pyridyl, R = CH_3

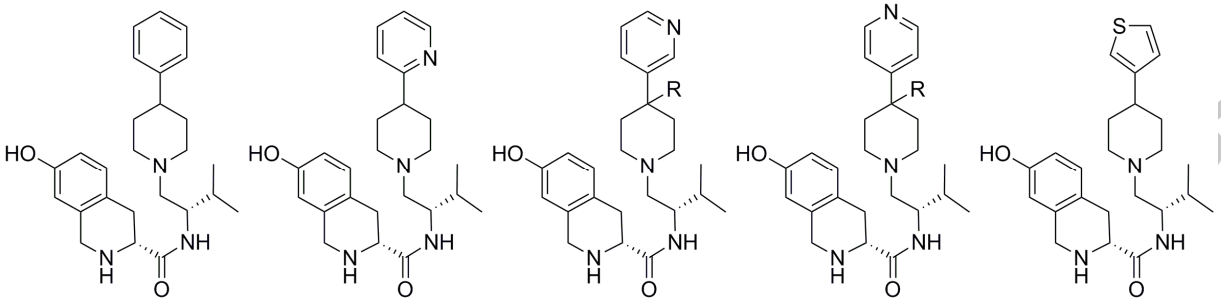
[a] Reagents: a) (1) *N*-Boc-L-valine, EDC•HCl, HOBt, Et_3N, CH_3CN, (2) HCl, (3) BH_3, THF; b) (1) 7-hydroxy-Boc-D-Tic, EDC•HCl, HOBt, Et_3N, CH_2Cl_2, (2) 4M HCl in dioxane.

3. Results and Discussion

In order to determine the effect of KOR potency and selectivity by replacing the phenyl ring in previously reported **1** with a 2-, 3-, or 4-pyridyl group or a thiophene ring, compounds **2**, **3a–b**, **4a–b**, and **5** were synthesized and first evaluated at 10 μM for intrinsic activity in the [^{35}S]GTPγS binding assay at all three opioid receptors. As none of these compounds displayed measurable intrinsic activity at this concentration, the compounds were evaluated for antagonist potency and selectivity at the opioid receptors. Compounds **3b** and **4b** were synthesized and tested for their KOR [^{35}S]GTPγS binding potency in order to determine the effect of adding a 4-methyl group to **3a** and **4a**.

Compounds **2**, **3a**, and **4a** with K_e values at the KOR of 0.43, 0.51, and 0.51 nM, respectively, were highly potent KOR antagonists but were slightly less potent than **1** which had a K_e = 0.051 nM at the KOR (Table 1). Similar to **1**, all three compounds were highly selective for the KOR relative to the DOR. Compounds **2**, **3a**, and **4a** were 33-, 89-, and 112-fold selective for the KOR relative to the MOR, compared to 77-fold selectivity for KOR relative to MOR for **1**.

Table 1. Inhibition of agonist-stimulated [^{35}S]GTPγS binding in cloned human μ, δ, and κ opioid receptors



1 **2** **3a**, R = H; **3b**, R = CH_3 **4a**, R = H; **4b**, R = CH_3 **5**

Compound	K_e (nM)[a]			μ/κ	δ/κ
	μ, DAMGO	δ, DPDPE	κ, U69,593		
JDTic	25 ± 4	74 ± 2	0.02 ± 0.01	1,250	3,800
1	3.96 ± 1.1	281 ± 44	0.051 ± 0.01	77	5,510
2	14.5 ± 2.6	590 ± 130	0.43 ± 0.12	33	1,372
3a	45.4 ± 15	1140 ± 70	0.51 ± 0.08	89	2,235
3b	49.2 ± 14	>3000	0.18 ± 0.01	273	>16,700
4a	57.1 ± 15	690 ± 250	0.51 ± 0.14	112	1,353
4b	154 ± 56	2,510 ± 490	1.75 ± 0.65	88	1,434
5	3.93 ± 0.73	118 ± 27	0.12 ± 0.02	33	983

[a] K_e values are the mean ± SEM of at least three independent experiments performed in duplicates.

The addition of a 4-methyl substituent to **3a** to give **3b** resulted in a significant increase in both KOR potency and selectivity for the KOR relative to both the MOR and DOR. Compound **3b** has a K_e = 0.18 nM at the KOR and is 273- and >16,700-fold selective for the KOR relative to the MOR and DOR, respectively, whereas compound **3a** has a K_e = 0.51 nM at the KOR and has 89- and 2,235-fold selectively for KOR relative to MOR and DOR, respectively. Compound **4b** has a K_e = 1.75 nM at the KOR and is 88- and 1,434 fold selective for the KOR relative to the MOR and DOR, respectively.

Compound **5**, which has a thiophene ring in place of a phenyl ring in **1** has a K_e value of 0.12 nM at KOR and is 32.5- and 983-fold selective for KOR relative to the MOR and DOR. Thus, **5** is less potent and less selective than **1**.

In order to determine if the compounds would be predicted to cross the blood/brain barrier, their topological polar surface area (TPSA), clogP, and derived logBB values were calculated and compared to JDTic and **1** (Table 2). In general, CNS compounds that have TPSA values less than 76 Å^2[9], clogP values in the range of 2-4[10], and derived logBB values greater than -1[11] are predicted to cross the blood/brain barrier. Compounds **3a–b** and **4a–b** have TPSA values of 77.49 Å^2 compared to 84.83 Å^2 for JDTic. Compound **5** has a TPSA value of 64.6, which is identical to that of **1**. JDTic, **3a–b, 4a–b** and **5** have clogP values in the range of 2–4. All of the compounds have logBB values greater than -1 and thus, would be predicted to penetrate the brain. In a study of marketed CNS drugs Wager and co-workers determined that 74% of CNS drugs displayed a CNS MPO (Multi Parameter Optimization) score greater than or equal to four.[12] All of the compounds in this study had a CNS MPO score greater than or equal to four. JDTic and **1** had CNS MPO scores of 3.1 and 3.8, respectively.

Table 2. Calculated physiochemical data for compounds **2**, **3a–b**, **4a–b**, and **5** compared to JDTic and compound **1**

Compound	TPSA	cLogP	ClarkBB	CNS MPO
JDTic	84.83	3.60	-0.57	3.1
1	64.60	3.53	-0.28	3.8
2	77.49	2.56	-0.62	4.0
3a	77.49	2.42	-0.64	4.2
3b	77.49	2.64	-0.61	4.1
4a	77.49	2.40	-0.64	4.2
4b	77.49	2.63	-0.61	4.1
5	64.60	3.45	-0.29	4.1

4. Conclusion

Compounds **2**, **3a–b**, **4a–b**, and **5**, which have either a pyridine or thiophene ring in place of the phenyl groups in JDTic analogs were synthesized and evaluted for their [^{35}S] GTPγS binding potency at the MOR, DOR, and KOR. All of the compounds had subnanomolar K_e values at the KOR in the [^{35}S] GTPγS binding assay except **4b** which has a K_e = 1.75 nM at the KOR. Compounds **3a–b** and **4a–b** were as selective or more selective for the KOR relative to the MOR than previously reported **1**. All of the compounds were highly selective for the KOR relative to the DOR. Compound **3b** with a K_e = 0.18 nM at the KOR with 273- and >16,700-fold selectively for the KOR relative to the MOR and DOR respectively, had the best overall antagonist potency and selectivity in the [^{35}S] GTPγS binding assays. The calculated logBB values suggest that the compounds will penetrate the blood-brain barrier and the calculated CNS MPO values are above 4 and thus are in the range for potential useful CNS drugs.

5. Materials and Methods

Melting points were determined using a MEL-TEMP II capillary melting point apparatus and are uncorrected. Nuclear magnetic resonance (^{1}H NMR and ^{13}C NMR) spectra were obtained on a Varian Avance DPX-300 MHz NMR spectrometer or a Bruker Unity Inova 500 MHz NMR spectrometer. Chemical shifts are reported in parts per million (ppm) with reference to internal solvent. Mass spectra (MS) were run on a Perkin-Elmer Sciex API 150 EX mass spectrometer equipped with APCI (atmospheric pressure chemical ionization) or ESI (turbospray) sources or on a Hewlett Packard 5989A instrument by electron impact. Elemental analyses were performed by Atlantic Microlab Inc., Atlanta, GA. 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 or using EM

Science silica gel 60A (230-400 mesh). Solvent system: CMA80=80:18:2 $CHCl_3$:MeOH:conc. NH_4OH. 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.

5.1. (3*R*)-7-Hydroxy-*N*-{(1*S*)-2-methyl-1-[(4-pyridin-2-ylpiperidin-1-yl)methyl]propyl}-1,2,3,4-tetrahydroisoquinoline-3-carboxamide (2) Trihydrochloride

To a solution of **13a** (200 mg, 0.56 mmol) in dichloromethane (25 mL) was added (3*R*)-2-(tert-butoxycarbonyl)-7-hydroxy-1,2,3,4-tetrahydroisoquinoline-3-carboxylic acid (7-hydroxy-Boc-D-Tic) (176 mg, 0.60 mmol) and triethylamine (0.35 mL, 0.6 mmol) followed by 1-ethyl-3-(3-dimethylaminopropyl)carbodiimide hydrochloride (EDC•HCl) (230 mg, 1.2 mmol) and hydroxybenzotriazole (HOBt) (8.1 mg, 0.06 mmol). The reaction mixture was stirred overnight at ambient temperature and then treated with 20 mL of saturated sodium bicarbonate and extracted with 25 mL dichloromethane. The combined organic layers were dried over sodium sulfate, filtered and evaporated. The resulting residue was subjected to chromatography on silica eluting with CMA 80-chloroform (1:1) provided 225 mg (77%) of the Boc-intermediate as a white solid. This material was dissolved in acetonitrile and treated with HCl (4M in dioxane, 1 mL) and stirred for 5 min. The solvent was evaporated to afford a white precipitate which was subjected to chromatography on silica eluting with CMA 80-chloroform (1:1) to provide 60 mg (33%) of **2** as the free base: ^{1}H NMR (300 MHz, METHANOL-d_4) δ 8.81 (d, *J* = 5.27 Hz, 1H), 8.61 (s, 1H), 8.10 (d, *J* = 8.10 Hz, 1H), 8.00 (d, *J* = 6.59 Hz, 1H), 7.13 (d, *J* = 8.48 Hz, 1H), 6.78 (dd, *J* = 2.45, 8.48 Hz, 1H), 6.68 (d, *J* = 2.26 Hz, 1H), 4.18–4.50 (m, 5H), 3.73 (br. s., 1H), 3.04–3.59 (m, 7H), 2.68–2.93 (m, 1H), 2.44–2.66 (m, 1H), 2.21–2.41 (m, 2H), 1.81–1.99 (m, 1H), 1.06 (s, 6H); ^{13}C NMR (75 MHz, METHANOL-d_4) δ 171.3, 159.2, 158.1, 148.2, 143.3, 131.2, 129.7, 126.9, 126.5, 121.9, 117.0, 113.7, 61.7, 57.9, 55.7, 52.4, 50.6, 45.5, 39.3,

32.3, 29.9, 29.0, 28.9, 19.9, 18.3; MS (ESI) *m/z* 423.5 $(M+H)^+$. A solution of the free base in CH_2Cl_2 was treated with HCl (2 M in ether) to afford the trihydrochloride salt (**2**•3HCl): mp 225–229 °C (dec); $[\alpha]^{25}_D$ = +69.2 (*c* 1.08, MeOH). Anal. Calcd for $C_{25}H_{37}Cl_3N_4O_2$•$2H_2O$: C, 52.87; H, 7.28; N, 9.86. Found: C, 52.95; H, 7.21; N, 10.12.

5.2. (3*R*)-7-Hydroxy-*N*-{(1*S*)-2-methyl-1-[(4-pyridin-3-ylpiperidin-1-yl)methyl]propyl}-1,2,3,4-tetrahydroisoquinoline-3-carboxamide (3a) Trihydrochloride

Compound **3a** was prepared according to procedure analogous to that of **2** from (2*S*)-3-methyl-1-(4-pyridin-3-ylpiperidin-1-yl)butan-2-amine (**13b**) (200 mg, 0.56 mmol), which in turn was prepared from amine **9b** according to procedure analogous to that of **13a**, to afford 109 mg (60%) of the **3a** free base. The free base was converted to the trihydrochoride salt (**3a**•3HCl): ^{1}H NMR (300 MHz, METHANOL-d_4) δ 8.90 (s, 1H), 8.81 (d, *J* = 5.65 Hz, 1H), 8.61–8.74 (m, 1H), 8.11 (dd, *J* = 5.93, 8.01 Hz, 1H), 7.11 (d, *J* = 8.29 Hz, 1H), 6.75 (dd, *J* = 2.26, 8.29 Hz, 1H), 6.67 (d, *J* = 2.07 Hz, 1H), 4.25–4.48 (m, 4H), 4.17 (d, *J* = 11.68 Hz, 1H), 3.70 (d, *J* = 11.49 Hz, 1H), 2.99–3.55 (m, 8H), 2.57–2.77 (m, 1H), 2.33–2.56 (m, 1H), 2.17 (d, *J* = 9.42 Hz, 2H), 1.77–2.01 (m, 1H), 1.03 (t, *J* = 5.75 Hz, 6H); ^{13}C NMR (126 MHz, METHANOL-d_4) δ 171.2, 158.0, 146.8, 145.8, 141.7, 141.4, 131.2, 129.7, 128.7, 121.9, 116.9, 113.7, 61.5, 57.8, 56.1, 52.8, 50.5, 45.4, 37.9, 32.3, 30.0, 29.9, 19.9, 18.4; MS (ESI) *m/z* 423.5 $(M+H)^+$; mp 223–227 °C; $[\alpha]^{25}_D$ = +70.0 (*c* 0.95, MeOH). Anal. Calcd for $C_{25}H_{37}Cl_3N_2O_2$•$1.5H_2O$: C, 53.72; H, 7.21; N, 10.02. Found: C, 53.57; H, 7.37; N, 9.83.

5.3. (3*R*)-7-Hydroxy-*N*-{(1*S*)-2-methyl-1-[(4-methyl-4-pyridin-3-ylpiperidin-1-yl)methyl]propyl}-1,2,3,4-tetrahydroisoquinoline-3-carboxamide (3b) Trihydrochloride

Compound **3b** was prepared according to procedure analogous to that of **2** from amine **13e** (117 mg, 0.45 mmol) to afford the **3b** freebase: ^{1}H NMR (300 MHz, CHLOROFORM-d) δ 8.50–8.65 (m, 1H), 8.43 (dd, *J* = 1.32, 4.71 Hz, 1H), 7.54–7.71 (m, 1H), 7.19–7.33 (m, 1H), 7.11 (d, *J* = 9.42 Hz, 1H),

6.82–6.97 (m, 1H), 6.60 (dd, *J* = 2.35, 8.19 Hz, 1H), 6.38–6.51 (m, 1H), 4.03–4.18 (m, 1H), 3.71–3.81 (m, 2H), 3.38 (dd, *J* = 5.27, 10.17 Hz, 1H), 2.94 (dd, *J* = 5.27, 16.39 Hz, 1H), 2.20–2.81 (m, 7H), 1.99–2.19 (m, 2H), 1.67–1.92 (m, 3H), 1.20–1.25 (m, 3H), 0.79–0.99 (m, 6H); ^{13}C NMR (75 MHz, CHLOROFORM-d) δ 173.2, 155.2, 147.6, 146.7, 144.3, 136.9, 133.9, 130.4, 125.0, 123.5, 114.1, 112.4, 60.0, 56.6, 50.3, 49.9, 47.5, 36.4, 36.2, 35.1, 31.2, 29.6, 19.1, 17.9. The free base was converted to the trihydrochloride salt affording 62.5 mg (25% over two steps) **3b**•3HCl as a white powder: MS (ESI) *m*/*z* 437.7 $(M+H)^+$, mp 215–219 °C (fusion), $[\alpha]^{25}_D$ +69. (*c* 0.10, CH_3OH). Anal. Calcd for $C_{26}H_{39}Cl_3N_4O_2$•H_2O: C, 53.66; H, 7.45; N, 9.63. Found: C, 53.49; H, 7.41; N, 9.50.

5.4. (3*R*)-7-Hydroxy-*N*-{(1*S*)-2-methyl-1-[(4-pyridin-4-ylpiperidin-1-yl)methyl]propyl}-1,2,3,4-tetrahydroisoquinoline-3-carboxamide (4a) Trihydrochloride

Compound **4a** was prepared according to procedure analogous to that of **2** from amine **13c** (280 mg, 1.13 mmol) to afford 113 mg (24%) of **4a** as the free base: ^{1}H NMR (300 MHz, METHANOL-d_4) δ 8.85 (d, *J* = 6.59 Hz, 2H), 8.12 (d, *J* = 6.59 Hz, 2H), 7.13 (d, *J* = 8.48 Hz, 1H), 6.78 (dd, *J* = 2.45, 8.29 Hz, 1H), 6.68 (d, *J* = 2.26 Hz, 1H), 4.30–4.45 (m, 4H), 4.23 (br. s., 1H), 3.71 (br. s., 1H), 3.04–3.54 (m, 7H), 2.63–2.87 (m, 1H), 2.37–2.61 (m, 1H), 2.08–2.32 (m, 2H), 1.91 (d, *J* = 6.59 Hz, 1H), 1.06 (t, *J* = 6.40 Hz, 6H); ^{13}C NMR (75 MHz, METHANOL-d_4) δ 171.3, 166.5, 158.1, 143.0, 131.2, 129.6, 127.3, 121.9, 117.0, 113.7, 61.7, 57.9, 56.1, 52.7, 50.5, 45.5, 40.9, 32.3, 29.9, 29.6, 29.5, 19.9, 18.3; MS (ESI) *m*/*z* 423.4 $(M+H)^+$. The free base was converted to the trihydrochloride salt (**4a**•3HCl): mp 238–241 °C (dec); $[\alpha]^{25}_D$ = +60.0 (*c* 0.98, MeOH). Anal. Calcd for $C_{25}H_{37}Cl_3N_4O_2$•H_2O: C, 54.60; H, 7.15; N, 10.19. Found: C, 54.31; H, 7.16; N, 9.91.

5.5. (3*R*)-7-Hydroxy-*N*-{(1*S*)-2-methyl-1-[(4-methyl-4-pyridin-4-ylpiperidin-1-yl)methyl]propyl}-1,2,3,4-tetrahydroisoquinoline-3-carboxamide (4b) Trihydrochloride

Compound **4b** was prepared according to procedure analogous to that of **2** from amine **13f** (78 mg, 0.30 mmol) to afford the **4b** freebase: ^{1}H NMR (300 MHz, METHANOL-d_4) δ 8.44 (dd, *J* = 1.60, 4.62 Hz, 2H), 7.43 (dd, *J* = 1.60, 4.62 Hz, 2H), 6.91 (d, *J* = 8.10 Hz, 1H), 6.52–6.68 (m, 1H), 6.48 (br. s., 1H), 3.79–4.16 (m, 3H), 3.44–3.65 (m, 1H), 2.72–3.00 (m, 2H), 2.63 (br. s., 1H), 2.19–2.56 (m, 5H), 1.94–2.17 (m, 2H), 1.65–1.91 (m, 3H), 1.05–1.35 (m, 4H), 0.75–1.04 (m, 6H); ^{13}C NMR (75 MHz, METHANOL-d_4) δ 175.4, 161.2, 156.8, 150.1, 137.3, 130.8, 125.6, 123.2, 114.9, 113.2, 61.4, 58.0, 52.5, 51.3, 51.2, 47.7, 37.5, 37.2, 37.1, 32.4, 32.1, 19.9, 17.9. The free base was converted to the trihydrochloride salt (**4b**•3HCl) affording 19.2 mg (11% over two steps) of a white powder: MS (ESI) *m*/*z* 437.5 $(M+H)^+$; mp 220–224 °C (fusion); $[\alpha]^{25}_{D}$ +75. (*c* 0.21, CH_3OH). Anal. Calcd for $C_{26}H_{39}Cl_3N_4O_2$•3.25H_2O: C, 51.66; H, 7.59; N, 8.85. Found: C, 51.88; H, 7.42; N, 9.27.

5.6 (3*R*)-7-Hydroxy-*N*-[(1*S*)-2-methyl-1-{[4-(3-thienyl)piperidin-1-yl]methyl}propyl]-1,2,3,4-tetrahydroisoquinoline-3-carboxamide (5)

Compound **5** was prepared according to procedure analogous to that of **2** from amine **13d** (680 mg, 2.7 mmol) to afford the **5** free base as a viscous, colorless oil (220 mg, 19%): ^{1}H NMR (300 MHz, CHLOROFORM-d) δ 7.35 (br. s., 1H), 7.14 (dd, *J* = 2.83, 4.71 Hz, 1H), 6.68–6.90 (m, 3H), 6.53 (d, *J* = 7.91 Hz, 1H), 6.39 (br. s., 1H), 4.12 (br. s., 1H), 3.62 (br. s., 2H), 3.21–3.45 (m, 2H), 3.06 (br. s., 1H), 2.84 (t, *J* = 15.16 Hz, 2H), 2.29–2.67 (m, 4H), 2.19 (br. s., 1H), 1.59–2.01 (m, 5H), 0.86 (d, *J* = 6.03 Hz, 6H); ^{13}C NMR (75 MHz, CHLOROFORM-d) δ 172.5, 154.1, 144.8, 135.5, 129.3, 125.6, 124.6, 123.7, 118.2, 113.1, 111.3, 58.4, 55.8, 54.3, 51.4, 48.7, 46.7, 35.5, 30.4, 28.9, 18.2, 17.0. The free base was converted to the trihydrochloride salt (**5**•3HCl): MS (ESI) *m*/*z* 428.2 $(M+H)^+$; mp 196–200 °C (fusion); $[\alpha]^{25}_{D}$ = +75.2 (*c* 1.0, MeOH). Anal. Calcd for $C_{24}H_{36}Cl_2N_3O_2S$•H_2O: C, 55.11; H, 7.23; N, 8.03. Found: C, 55.09; H, 7.00; N, 7.80.

5.7. *tert*-Butyl 4-thiophen-3-ylpiperidine-1-carboxylate (8d)

A mixture of **6** (3.2 g, 9.6 mmol), thiophene-3-boronic acid (1.72 g, 13.5 mmol), tetrakis(triphenylphosphine)palladium (1.1 g, 0.96 mmol), cesium carbonate (9.4 g, 28.6 mmol), dimethoxyethane (34 mL) and water (16 mL) was stirred at 80 °C under nitrogen overnight. The cooled mixture was partitioned between water and EtOAc. The aqueous was further extracted with EtOAc (100 mL). The combined organic layers were dried (Na_2SO_4), filtered and evaporated to obtain intermediate *tert*-butyl 4-thiophen-3-yl-3,6-dihydropyridine-1(2*H*)-carboxylate **7d**: ^{1}H NMR (300 MHz, CHLOROFORM-d) δ 7.10–7.26 (m, 2H), 7.05 (br. s., 1H), 5.98 (br. s., 1H), 3.99 (d, *J* = 2.64 Hz, 2H), 3.55 (t, *J* = 5.75 Hz, 2H), 2.44 (br. s., 2H), 1.36–1.46 (m, 9H). Intermediate **7d** was hydrogenated with palladium hydroxide on carbon (1.0 g, 20 wt%) in ethanol under H_2 (50 psi) at room temperature for 2 h. The solids were filtered and solvent evaporated to provide 1.25 g (35% over 2 steps) of **8d**: ^{1}H NMR (300 MHz, CHLOROFORM-d) δ 7.17–7.23 (m, 1H), 6.80–6.97 (m, 2H), 3.92–4.27 (m, 2H), 2.71 (d, *J* = 12.06 Hz, 3H), 1.82 (br. s., 2H), 1.52 (br. s., 2H), 1.36–1.43 (m, 9H).

5.8. 4-Thiophen-3-ylpiperidine (9d)

A solution of **8d** (1.25 g, 4.67 mmol) in acetonitrile (20 mL) was treated with 4M HCl in dioxane (4 mL) and the mixture stirred for 1 h at room temperature. The solvent was evaporated to obtain 0.86 g (90%) of **9d** as the hydrochloride salt: ^{1}H NMR (300 MHz, DMSO-d_6) δ 8.66–9.67 (m, 2H), 7.40–7.59 (m, 1H), 7.14–7.28 (m, 1H), 6.97–7.11 (m, 1H), 3.14–3.39 (m, 2H), 2.81–3.06 (m, 3H), 1.93–2.12 (m, 2H), 1.67–1.91 (m, 2H).

5.9. 3-(4-Methylpiperidin-4-yl)pyridine (9e)

A solution of **12a** (3.95 g, 19.3 mmol) in THF (50 mL) was treated with BH_3•SMe_2 (10 mL, 100 mmol) at 0°C, then heated to reflux for 12 h. The reaction mixture was quenched by addition of EtOAc, then methanol. The concentrated residue was dissolved in aq. HCl (2.0 M, 10 mL). The pH was adjusted with NaOH (2 M, 12 mL) and the resulting aqueous layer was extracted with EtOAc (3 x 50 mL). The

combined organics were washed with brine, dried (Na_2SO_4) and concentrated. The resulting residue was subjected to chromatography on silica gel eluting with a gradient up to 100% CMA80 in CH_2Cl_2 to afford 567 mg (17%) of piperidine **9e**: ^{1}H NMR (300 MHz, CHLOROFORM-d) δ 8.54–8.67 (m, 1H), 8.44 (d, *J* = 3.77 Hz, 1H), 7.56–7.71 (m, 1H), 7.18–7.33 (m, 1H), 3.57 (br. s., 1H), 2.89–3.04 (m, 1H), 2.74–2.89 (m, 1H), 1.52–2.70 (m, 7H), 1.28 (d, *J* = 7.72 Hz, 2H).

5.10. 4-(4-Methylpiperidin-4-yl)pyridine (9f)

Compound **9f** was prepared according to procedure analogous to that of **9e** from glutarimide **12b** (652 mg, 3.2 mmol) to afford 100 mg (18%) of piperidine **9f**: ^{1}H NMR (300 MHz, $CDCl_3$) δ 8.39–8.63 (m, 2H), 7.25 (dd, *J* = 1.9, 4.1 Hz, 2H), 2.86–3.02 (m, 2H), 2.71–2.86 (m, 2H), 1.93–2.12 (m, 2H), 1.60–1.79 (m, 2H), 1.17–1.31 (m, 3H); ^{13}C NMR (75 MHz, $CDCl_3$) δ 158.4, 149.7, 121.1, 42.6, 37.2, 36.6, 28.7; MS (ESI) *m*/*z* 177.3 $(M+H)^+$.

5.11. 4-Methyl-2,6-dioxo-4-(pyridin-3-yl)piperidine-3,5-dicarbonitrile (11a)

A solution of 3-acetylpyridine (**10a**) (10.0 g, 82.5 mmol, ethyl cyanoacetate (9.33 g, 82.5 mmol) and ammonium acetate (1.3 g, 16.9 mmol) was refluxed in glacial acetic acid (4.0 mL) and benzene (16.5 mL) with a Dean-Stark apparatus for 12 h. Ethyl acetate was added to the cooled solution. The solution was washed with aq. $NaHCO_3$ then brine, dried (Na_2SO_4) and concentrated. The residue was eluted through a plug of silica gel using ethyl acetate to afford 16.32 g (92%) of the Knoevenagel product. The oil was combined with 2-cyanoacetamide (7.36 g, 87.5 mmol) in a solution of NaOH (3.5 g, 87.5 mmol) in ethanol (60 mL). Within 5 minutes, a clear solution had formed which was left to stir for 12 h, after which aq. $NaHSO_4$ (1 M, 90 mL) was added. The resulting solids were collected by filtration and the mother liquor was concentrated to approximately half the original volume of kept in a refrigerator overnight. A second filtration yielded more solids, which were combined with the first crop and dried under vacuum to afford 11.6 g (55% over two steps) of **11a**: ^{1}H NMR (300 MHz, DMSO-d_6) δ

12.40 (br. s., 1H), 8.48–9.02 (m, 2H), 8.08 (d, J = 6.59 Hz, 1H), 7.57 (dd, J = 4.71, 7.91 Hz, 1H), 5.42 (s, 2H), 1.72 (br. s., 3H).

5.12. 4-Methyl-2,6-dioxo-4-(pyridin-4-yl)piperidine-3,5-dicarbonitrile (11b)

A solution of 4-acetylpyridine (**10b**) (10.0 g, 82.5 mmol), ethyl cyanoacetate (9.33 g, 82.5 mmol) and ammonium acetate (1.3 g, 16.9 mmol) was refluxed in glacial acetic acid (4.0 mL) and benzene (16.5 mL) with a Dean-Stark apparatus for 12 h. Ethyl acetate was added to the cooled solution. The solution was washed with aq. $NaHCO_3$ then brine, dried (Na_2SO_4) and concentrated. The residue was eluted through a plug of silica gel using ethyl acetate to afford 13.76 g of an amber oil. The oil was combined with cyanoacetamide (5.35 g, 63.6 mmol) in methanol (200 mL) at 0 °C. Ammonia gas was bubbled in to the solution until it was saturated, then the dark red solution was left to warm to room temperature and stir 1 week. The solution was brought to reflux for 1 h, at which point the solution became cloudy. The solution was concentrated to a thick oil which was dissolved in EtOAc. Diethyl ether was slowly added, causing a precipitate to form. Filtration afforded 8.81 g (54%) of **11b** as a pale yellow powder. ^{1}H NMR (300 MHz, DMSO-d_6) δ 9.39 (d, J = 4.1 Hz, 1H), 8.50 (ddd, J = 1.6, 3.2, 4.6 Hz, 2H), 7.20–7.60 (m, 2H), 4.31–4.74 (m, 1H), 1.28–1.79 (m, 3H); MS (ESI) m/z 253.3 $(M–H)^-$.

5.13. 4-Methyl-4-(pyridin-3-yl)piperidine-2,6-dione (12a)

Sulfuric acid (6 mL) was slowly added to a slurry of **11a** (11.6 g, 11.8 mmol) in water (6 mL). The resulting solution was refluxed for 12 h, cooled, quenched with 50% NaOH (6 mL), diluted with methanol (40 mL) then CH_2Cl_2 (160 mL). The white solids were filtered and washed with 10% methanol in CH_2Cl_2. The orange filtrate was concentrated to afford 11.03 g of crude material, which was combined with urea (7.1 g, 0.12 mol) and heated as a melt for 3 h until no further gas evolution was observed. The mixture was partitioned between aq. $NaHCO_3$ and EtOAc, and the resulting emulsion was filtered through Celite. The aqueous layer was further extracted with EtOAc. The combined organic

layers were dried (Na_2SO_4) and concentrated. The resulting residue was eluted through a plug of silica gel using 5–7.5% MeOH in CH_2Cl_2 as the eluent. The concentrated residue, upon trituration with EtOAc, afforded 3.95 g (42% over two steps) of **12a** as a solid: ^{1}H NMR (300 MHz, DMSO-d_6) δ 10.80 (br. s., 1H), 8.63 (d, *J* = 2.64 Hz, 1H), 8.36–8.52 (m, 1H), 7.74–7.90 (m, 1H), 7.37 (dd, *J* = 4.71, 8.10 Hz, 1H), 2.98–3.17 (m, 1H), 2.75–2.94 (m, 1H), 1.33 (br. s., 1H).

5.14. 4-Methyl-4-(pyridin-4-yl)piperidine-2,6-dione (12b)

Sulfuric acid (1.5 mL) was slowly added to a slurry of **11b** (3.0 g, 11.8 mmol) in water (1.5 mL). The resulting solution was refluxed for 12 h, cooled, quenched with 50% NaOH (1.5 mL), diluted with methanol (10 mL) then CH_2Cl_2 (40 mL). The white solids were filtered and washed with 10% methanol in CH_2Cl_2. The orange filtrate was concentrated to afford 2.23 g (85%) of 3-methyl-3-(pyridin-4-yl)pentanedioic acid. ^{1}H NMR (300 MHz, CD_3OD) δ 8.23 – 8.62 (m, 2H), 7.30 – 7.62 (m, 2H), 2.70 – 3.01 (m, 4H), 1.58 (br. s., 3H). The 3-methyl-3-(pyridin-4-yl)pentanedioic acid (2.23 g, 10.0 mmol) was combined with urea (2.0 g, 33 mmol) and heated as a melt for 1 h. The resulting material was subjected to chromatography on silica gel. Although the product had R_f =0.3 in EtOAc, the solubility in pure EtOAc was limited, so a gradient up to 10% methanol in EtOAc was used to afford 652 mg (32%) of **12b** as a white solid. ^{1}H NMR (300 MHz, $CDCl_3$) δ 8.59 (dd, *J* = 1.60, 4.4 Hz, 2H), 7.25 (dd, *J* = 1.6, 4.4 Hz, 2H), 2.97–3.19 (m, 2H), 2.80 (d, *J* = 17.0 Hz, 2H), 1.45 (s, 3H); MS (ESI) *m/z* 205.3 $(M+H)^+$.

5.15. (2*S*)-3-methyl-1-(4-pyridin-2-ylpiperidin-1-yl)butan-2-amine (13a)

A solution of **9a** (4.66 g, 19.8 mmol) in acetonitrile (150 mL) was treated with *N*-Boc-L-valine (4.55 g, 21.0 mmol), HBTU (7.98 g, 21.0 mmol) and triethylamine (8.4 g, 84 mmol). The reaction mixture was stirred overnight at ambient temperature then evaporated. The residue was partitioned between sat. aq. $NaHCO_3$ (100 mL) and EtOAc (100 mL). The aqueous was extracted further with EtOAc (2 x 100 mL). The combined organic layer was dried (Na_2SO_4) then evaporated to obtain the

crude Boc intermediate which was subjected to silica gel chromatography eluting with a gradient up to 10% MeOH in CH_2Cl_2: MS (ESI) *m/z* 362.3 $(M+H)^+$. The purified Boc intermediate was dissolved in acetonitrile (100 mL), treated with 4M HCl in dioxane (20 mL) and stirred for 2 h. The reaction was evaporated to obtain a solid which was subjected to chromatography on silica gel using a gradient of 10-50% CMA80 in $CHCl_3$ to afford 5.5 g (83% over two steps) of the intermediate amide (2*S*)-3-methyl-1-oxo-1-(4-pyridin-2-ylpiperidin-1-yl)butan-2-amine as an off white solid: MS (ESI) *m/z* 262.2 $(M+H)^+$. A solution of the intermediate amide (5.5 g, 21 mmol) in THF (40 mL) at 0 °C was treated with borane dimethyl sulfide complex (3.3 mL, 33 mmol). The reaction mixture was warmed to ambient temperature then heated at reflux overnight. The reaction was cooled in an ice bath, treated with 20 mL methanol then warmed to ambient temperature over 1 h. The resulting solution was again cooled in an ice bath, treated with 2M HCl in ether (100 mL) and then heated at reflux for 1 h. The solvent was evaporated and the residue was subjected to chromatography on silica gel eluting with a gradient of CMA80 in $CHCl_3$ to afford 2.2 g (42%) of **13a** as a white solid.

5.16. (2*S*)-3-methyl-1-(4-pyridin-4-ylpiperidin-1-yl)butan-2-amine (13c)

Compound **13c** was prepared according to the general procedure of **13a** from the amine **9c** (1.42 g, 5.9 mmol) to afford 0.77 g (53% yield) of the desired **13c** free base: ^{1}H NMR (300 MHz, METHANOL-d_4) δ 8.42–8.52 (m, 2H), 7.35–7.46 (m, 2H), 3.21 (q, *J* = 7.35 Hz, 2H), 3.00 (d, *J* = 11.11 Hz, 1H), 2.42–2.77 (m, 3H), 2.19 (dt, *J* = 4.33, 10.83 Hz, 1H), 1.84–1.97 (m, 4H), 1.32 (t, *J* = 7.35 Hz, 2H), 0.98–1.11 (m, 6H).

5.17. (2*S*)-3-Methyl-1-[4-(thiophen-3-yl)piperidin-1-yl]butan-2-amine (13d)

A solution of 4-(thiophen-3-yl)piperidine **(9d)** (850 mg, 4.17 mmol) in CH_3CN (30 mL) was treated with *N*-Boc-L-valine(1.08 g, 5.0 mmol), HBTU (1.9 g, 5.0 mmol) and NEt_3 (1.5 g, 15 mmol). The reaction mixture was stirred overnight at ambient temperature. The concentrated residue was

partitioned between sat. aq. $NaHCO_3$ (15 mL) and EtOAc (15 mL). The aqueous layer was further extracted with EtOAc (2 x 15 mL). The combined organic layer was dried (Na_2SO_4), filtered, and evaporated. The residue was subjected to chromatography on silica gel eluting with 50% EtOAc in hexanes to afford the intermediate *tert*-butyl {(1*S*)-2-methyl-1-[(4-thiophen-3-ylpiperidin-1-yl)carbonyl]propyl}carbamate as a light yellow oil (1.34 g, 88%): ^{1}H NMR (300 MHz, CHLOROFORM-d) δ 7.20–7.38 (m, 1H), 6.89–7.07 (m, 2H), 5.40 (d, *J* = 9.04 Hz, 1H), 4.63–4.84 (m, 1H), 4.54 (br. s., 1H), 3.92–4.20 (m, 1H), 3.10–3.29 (m, 1H), 2.90 (t, *J* = 11.68 Hz, 1H), 2.65–2.80 (m, 1H), 1.85–2.18 (m, 3H), 1.52–1.77 (m, 1H), 1.45 (s, 9H), 0.82–1.07 (m, 7H). The Boc-intermediate was dissolved in CH_3CN (10 mL) then treated with HCl (4 N in dioxane, 10 mL). After 1 h at ambient temperature, the solution was evaporated to afford a quantitative yield (1.09 g) of the (2*S*)-3-methyl-1-oxo-1-(4-thiophen-3-ylpiperidin-1-yl)butan-2-amine hydrochloride salt: ^{1}H NMR (300 MHz, DMSO-d_6) δ 8.21 (d, *J* = 11.30 Hz, 3H), 7.38–7.58 (m, 1H), 7.21 (d, *J* = 2.83 Hz, 1H), 6.96–7.14 (m, 1H), 4.50 (d, *J* = 12.62 Hz, 1H), 4.27 (br. s., 1H), 4.04 (d, *J* = 10.36 Hz, 1H), 3.17 (t, *J* = 12.43 Hz, 1H), 2.91 (t, *J* = 10.46 Hz, 1H), 2.75 (t, *J* = 12.53 Hz, 1H), 1.81–2.14 (m, 3H), 1.30–1.78 (m, 2H), 0.83–1.09 (m, 6H); MS (ESI) *m/z* 267.2 $(M+H)^+$. The intermediate amide was dissolved in THF (25 mL) then treated with borane dimethyl sulfide complex (1.8 mL, 18 mmol) at 0 °C. The reaction mixture was warmed to ambient temperature and stirred overnight. The reaction mixture was refluxed for 2 h, cooled in ice, treated with methanol (20 mL) and stirred for 1 h at ambient temperature. The reaction mixture was cooled in ice and treated with 2M hydrogen chloride in ether (40 mL) then refluxed overnight. The solvent was evaporated and the resulting residue was subjected to chromatography on silica gel eluting with a gradient up to 30% CMA80 in $CHCl_3$ to afford 680 mg (75%) of **13d** as a white solid: ^{1}H NMR (300 MHz, CHLOROFORM-d) d 7.13–7.25 (m, 1H), 6.82–6.98 (m, 2H), 4.91 (br. s., 2H), 2.78–3.03 (m, 3H), 2.48–2.62 (m, 1H), 2.36–2.45 (m, 2H), 2.29 (dt, *J* = 2.54, 11.54 Hz, 1H), 1.53–2.07 (m, 6H),

1.03 (d, *J* = 6.78 Hz, 3H), 0.93 (d, *J* = 6.78 Hz, 3H); ^{13}C NMR (75 MHz, CHLOROFORM-d) d 146.9, 126.8, 125.3, 118.9, 59.7, 55.6, 54.4, 52.9, 37.4, 33.3, 32.9, 30.3, 19.3, 18.8; MS (ESI) *m*/*z* 253.2 $(M+H)^+$.

5.18. (2*S*)-3-Methyl-1-(4-methyl-4-pyridin-3-ylpiperidin-1-yl)butan-2-amine (13e)

A solution of **9e** (152 mg, 0.86 mmol) and Boc-L-valine (230 mg, 1.06 mmol) in acetonitrile (30 mL) was treated with EDC•HCl (340 mg, 1.8 mmol) and triethylamine (0.7 mL, 5 mmol) and stirred 12 h. The concentrated residue was partitioned between EtOAc and water. The organic layer was washed (aq. $NaHCO_3$ then brine) then dried (Na_2SO_4). The concentrated residue was eluted from silica gel using ethyl acetate to afford an oil which was dissolved in methanol (10 mL) and treated with HCl (12 N, 2 mL), stirred 1 h and then concentrated. The residue was subjected to chromatography on silica gel eluting with a gradient up to 50% CMA80 in CH_2Cl_2 to afford 201 mg (84% over two steps) of the intermediate amide: ^{1}H NMR (300 MHz, CHLOROFORM-d) δ 8.64 (br. s., 1H), 8.40–8.57 (m, 1H), 7.55–7.74 (m, 1H), 7.18–7.39 (m, 1H), 1.60–4.27 (m, 12H), 1.33 (d, *J* = 11.87 Hz, 3H), 0.83–1.11 (m, 6H); MS (ESI) *m*/*z* 276.5 $(M+H)^+$. A solution of the intermediate amide (201 mg, 0.73 mmol) in THF (3 mL) was treated with BH_3•THF (1 M, 10 mL) then heated to reflux overnight. The reaction mixture was quenched with methanol and concentrated. The residue was subjected to chromatography on silica gel eluting with 25% CMA80 in CH_2Cl_2 to afford 117 mg (62%) of the desired **13e**: ^{1}H NMR (300 MHz, CHLOROFORM-d) δ 8.55 (s, 1H), 8.39–8.50 (m, 1H), 7.95 (d, *J* = 7.91 Hz, 1H), 7.50 (dd, *J* = 5.93, 7.82 Hz, 1H), 4.56 (br. s., 2H), 3.58–3.72 (m, 1H), 2.73–2.96 (m, 1H), 2.49–2.73 (m, 2H), 2.21–2.50 (m, 3H), 1.95–2.21 (m, 2H), 1.54–1.94 (m, 3H), 1.15–1.42 (m, 3H), 0.72–1.11 (m, 6H).

5.19. (2*S*)-3-Methyl-1-[4-methyl-4-(pyridin-4-yl)piperidin-1-yl]butan-2-amine (13f)

Compound **13f** was prepared according to procedure analogous to that of **13e** from amine **9f** (100 mg, 0.57 mmol) to afford 78 mg (52% over three steps) of **13f**: ^{1}H NMR (300 MHz, $CDCl_3$) δ 8.53 (dd,

J = 2.3, 4.0 Hz, 2H), 7.25 (d, *J* = 3.2 Hz, 2H), 2.33–2.76 (m, 4H), 1.96–2.33 (m, 7H), 1.67–1.88 (m, 2H), 1.41–1.61 (m, 1H), 1.15–1.27 (m, 3H), 0.81–0.99 (m, 6H).

5.20. [^{35}S]GTPγS assay

The [^{35}S]GTPγS assays were conducted using the methods previously reported.[13]

5.21. Calculated pharmacokinetic properties

The topological polar surface area (TPSA) and calculated lipophilicity (clogP) values were calculated using the ChemAxon Instant JChem package. Predictions of the logarithm of the in vivo blood-brain ratio (logBB) were based on the Clark and Pickett model (Eq.3).[11, 14]

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Supplementary data

Supplementary data ssociated with this article can be found, in the online version, at http://dx.doi.org/10.1016/j.bmc.

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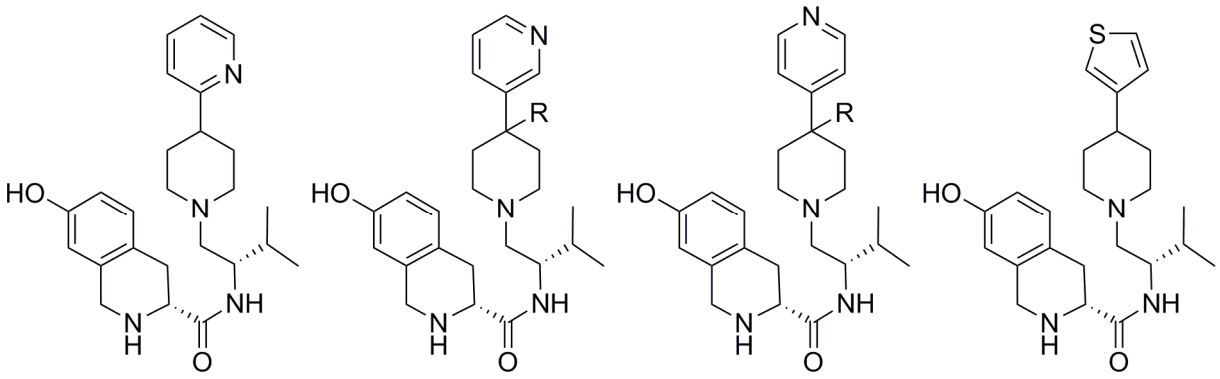
Design, synthesis, and pharmacological evaluation of JDTic analogs to examine the significance of replacement of the 3-hydroxyphenyl group with pyridine or thiophene bioisosteres

Chad M. Kormos, Moses G. Gichinga, Scott P. Runyon, James B. Thomas, S. Wayne Mascarella, Ann M. Decker, Hernán A. Navarro, and F. Ivy Carroll



2 **3a**, R = H; **3b**, R = CH_3 **4a**, R = H; **4b**, R = CH_3 **5**