Available online at www.sciencedirect.com
ScienceDirect
Bioorganic & Medicinal Chemistry Letters 18 (2008) 2860–2864
Bioorganic & Medicinal Chemistry Letters

2-Aminomethyl piperidines as novel urotensin-II receptor antagonists

Jian Jin,[a,*] Yonghui Wang,[b] Feng Wang,[c] Dongchuan Shi,[a] Karl F. Erhard,[b] Zining Wu,[c] Brian F. Guida,[c] Sarah K. Lawrence,[c] David J. Behm,[a] Jyoti Disa,[a] Kalindi S. Vaidya,[c] Christopher Evans,[a] Lynette J. McMillan,[c] Ralph A. Rivero,[b] Michael J. Neeb[a] and Stephen A. Douglas[a]

[a]*Cardiovascular and Urogenital Center of Excellence for Drug Discovery, GlaxoSmithKline, 709 Swedeland Road, King of Prussia, PA 19406, USA*
[b]*Oncology Center of Excellence for Drug Discovery, GlaxoSmithKline, 1250 South Collegeville Road, Collegeville, PA 19426, USA*
[c]*Molecular Discovery Research, GlaxoSmithKline, 1250 South Collegeville Road, Collegeville, PA 19426, USA*

Received 12 February 2008; revised 24 March 2008; accepted 31 March 2008
Available online 8 April 2008

Abstract—A series of 2-aminomethyl piperidines has been discovered as novel urotensin-II receptor antagonists. The synthesis, initial structure-activity relationships, and optimization of the initial hit that resulted in the identification of potent, cross-species active, and functional urotensin-II receptor antagonists such as **1a** and **11a** are described.

Human urotensin-II (hU-II), the most potent mammalian vasoconstrictor identified to date,[1] and its cognate receptor hUT (formerly known as the GPR-14 receptor) are proposed to be involved in the (dys)regulation of cardiorenal function,[2] and have been implicated in the etiology of numerous cardiorenal and metabolic diseases including hypertension,[3] heart failure,[4,5] atherosclerosis,[6] renal failure,[7] and diabetes.[8] The impressive pharmacological activity of U-II has stimulated a great deal of interest in developing small molecule UT modulators. A number of non-peptidic UT ligands have recently been reported.[9] Herein we describe the identification, synthesis, and initial structure-activity relationships (SAR) of a novel 2-aminomethyl piperidine series. Optimization of the series led to the identification of potent, competitive, and reversible UT antagonists such as **1a** and **11a** with excellent and broad cross-species functional activity.

High throughput screening (HTS) of the corporate compound collection using a fluorometric imaging plate reader (FLIPR) assay (measuring inhibition of hU-II-mediated $[Ca^{2+}]_i$-mobilization in HEK293 cells expressing human recombinant UT receptor)[10] led to the identification of **2**[11] as an antagonist with a pIC_{50} of 6.2 (Fig. 1). The compound also showed moderate hUT binding affinity with a pK_i of 6.4 in a [^{125}I]hU-II radioligand binding assay using HEK293 cell membranes stably expressing human recombinant UT receptors.[10] Subsequent early exploration of the left-hand side (LHS) of this hit quickly resulted in the identification of an α-aryl acetamide sub-series exemplified by **3a** (pK_i 6.3).[12] Despite the modest binding affinity, compounds

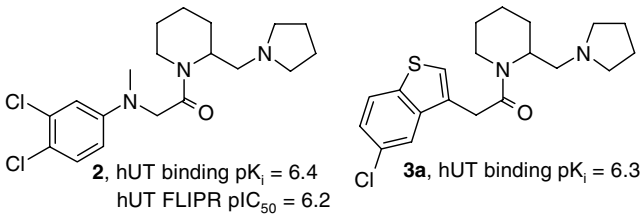
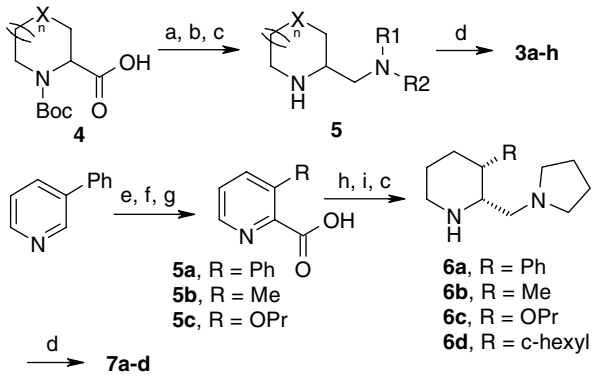


Figure 1. Structures of HTS hit **2** and α-aryl acetamide sub-series hit **3a**.

Keywords: Urotensin-II receptor antagonist; UT antagonist; 2-aminomethyl piperidine; Broad cross-species activity; Competitive, reversible, and functional antagonist.
* Corresponding author. Tel.: +1 610 270 4881; fax: +1 610 270 4490; e-mail: jian.jin@gsk.com

0960-894X/$ - see front matter © 2008 Elsevier Ltd. All rights reserved.
doi:10.1016/j.bmcl.2008.03.078



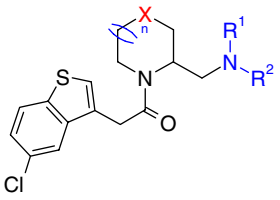
Scheme 1. Reagents and conditions: (a) amine, EDC, HOAt, CH_2Cl_2, rt; (b) 4 M HCl in dioxane, MeOH, rt; (c) $LiAlH_4$, THF, 0 °C–rt; (d) 2-(5-chlorobenzothiophen-3-yl)acetic acid, EDC, HOAt, CH_2Cl_2, rt; (e) *m*-CPBA, CH_2Cl_2, 0 °C–rt; (f) $(CH_3)_2NCOCl$, TMSCN, CH_2Cl_2, rt; (g) concd HCl and H_2SO_4, reflux; (h) pyrrolidine, EDC, HOAt, CH_2Cl_2, rt; (i) H_2, PtO_2, HOAc, rt.

2 and **3a** were considered as reasonable starting points for our hit-to-lead chemistry optimization aimed at improving potency via SAR exploration.

We first investigated the 2-aminomethyl piperidine region, also referred to as the diamine region. Custom diamines **5** were prepared from Boc-protected amino acids **4** via amide formation, deprotection, and $LiAlH_4$ reduction (Scheme 1). Subsequent coupling of diamines **5** with commercially available 2-(5-chlorobenzothiophen-3-yl)acetic acid produced the desired compounds **3a**–**h**.[13] To explore the effect of 3-substituents on the central piperidine ring, 3-phenylpyridine was first converted to the corresponding 2-carboxylic acid **5a** via *N*-oxide formation, installation of the 2-CN group,[14] and hydrolysis. Acid **5a** and commercially available **5b**–**c** were then converted to racemic cis-3-substituted-2-pyrrolidinylmethyl piperidines **6a**–**d** via amide formation, reduction of the pyridine ring, and subsequent amide reduction.[15] Standard amide coupling of **6a**–**d** with commercially available acids produced the desired compounds **7a**–**d** and **8a**–**j**.

For the 2-aminomethyl moiety, pyrrolidine (**3a**) was greater than 10-fold more potent compared to piperidine (**3b**) and acyclic analog (**3e**) (Table 1). Interestingly, unlike piperidine (**3b**), morpholine (**3c**) showed moderate binding affinity while *N*-methyl piperazine (**3e**) had no appreciable affinity. The SAR indicated that the size and the basicity of the 2-aminomethyl moiety were critical to UT binding. As for the central piperidine ring moiety, 6-membered ring (**3a**) was preferred compared to 5- and 7-membered rings (**3g** and **h**) while morpholine (**3f**) was tolerated. Additional substituents on the central piperidine ring were then explored. We were pleased to find that 3-substituents (**7a**–**d**) improved affinity with 3-phenyl (**7a**) being optimal—resulting in close to 100-fold affinity improvement compared to **3a** (Table 2). The 4- and 5-phenyl analogs (**7e** and **f**)[16] also had higher binding affinity compared to **3a**, but were less potent compared to 3-phenyl compound **7a**.

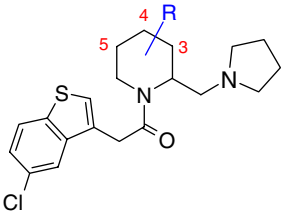
Table 1. SAR of the diamine region



Compound	*n*	X	NR^1R^2	hUT binding (pK_i)[a]
3a	1	CH_2	Pyrrolidin-1-yl	6.3
3b	1	CH_2	Piperidin-1-yl	5.1
3c	1	CH_2	Morpholin-4-yl	5.9
3d	1	CH_2	*N*-Methylpiperazin-1-yl	<5.1
3e	1	CH_2	*N*,*N*-Diethylamino	<5.1
3f	1	O	Pyrrolidin-1-yl	6.1
3g	0	CH_2	Pyrrolidin-1-yl	<5.1
3h	2	CH_2	Pyrrolidin-1-yl	5.7

[a] Mean of at least 3 determinations with standard deviation of <±0.3.

Table 2. Substitution on the central piperidine ring



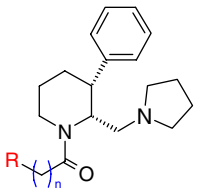
Compound	R	Relative stereochemistry	hUT binding (pK_i)[a]
3a	H	—	6.3
7a	3-Phenyl	*cis*	8.1
7b	3-Methyl	*cis*	6.7
7c	3-Propoxy	*cis*	7.5
7d	3-Cyclohexyl	*cis*	7.6
7e	4-Phenyl	*cis*	6.7
7f	5-Phenyl	*trans*	7.5

[a] Mean of at least 3 determinations with standard deviation of <±0.3.

After identifying **6a** as the optimal diamine, we next turned our attention to optimizing the left-hand side (LHS) moiety of the α-aryl acetamide sub-series. Compared to unsubstituted 3-benzothiophene (**8c**), 5-substituted-3-benzothiophenes (**7a**, **8a**–**b**) were 15- to 30-fold more potent, with 5-Cl (**7a**) and 5-Br (**8a**) being optimal (Table 3). Indole is less preferred (**8d** vs **8a**) and 1-naphthyl (**8e**) was 100-fold more potent than 2-naphthyl (**8f**). Monocyclic aromatic groups such as 3-thiophene (**8g**) and 3,4-dichlorophenyl (**8h**), the preferred LHS group in the α-amino acetamide sub-series (*vide infra*), had much lower binding affinity. As for the length of the carbon linker, one carbon was preferred over no or two carbons (**8e** vs **8i**, **8b** vs **8j**).

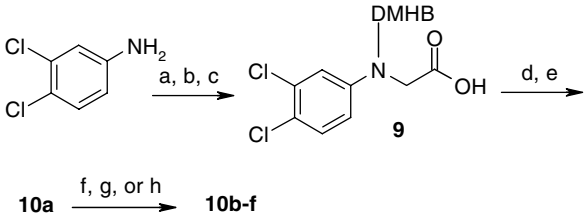
To efficiently explore the α-amino acetamide sub-series, a solid-phase synthetic route outlined in Scheme 2 was developed. Resin-bound α-amino acetic acid **9** was prepared via loading of 3,4-dichloroaniline onto commercially available 2,6-dimethoxy-4-polystyrene-benzyl-

Table 3. SAR of the LHS moiety of the α-aryl acetamide sub-series



Compound	R	*n*	hUT binding (pK_i)[a]
7a	5-Chlorobenzothiophen-3-yl	1	8.1
8a	5-Bromobenzothiophen-3-yl	1	8.1
8b	5-Methylbenzothiophen-3-yl	1	7.8
8c	Benzothiophen-3-yl	1	6.6
8d	5-Bromoindol-3-yl	1	6.2
8e	1-Naphthyl	1	7.1
8f	2-Naphthyl	1	5.1
8g	Thiophen-3-yl	1	<5.1
8h	3,4-Dichlorophenyl	1	5.6
8i	1-Naphthyl	0	<5.1
8j	5-Methylbenzothiophen-3-yl	2	5.5

[a] Mean of at least 3 determinations with standard deviation of <±0.3.

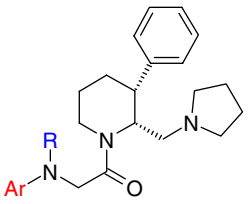


Scheme 2. Reagents and conditions: (a) 2,6-dimethoxy-4-polystyrenebenzyloxy-benzaldehyde, $Na(OAc)_3BH$, DIEA, 1% of HOAc in NMP, rt; (b) methyl bromoacetate, DIEA, NMP, 80 °C; (c) potassium trimethylsilanolate, THF, rt; (d) **6a**, PyBOP, DIEA, NMP, rt; (e) 50% of TFA in DCE, rt; (f) aldehyde, $Na(OAc)_3BH$, HOAc, DIEA, DCE, rt; (g) acyl chloride, TEA, CH_2Cl_2, rt; (h) $MeSO_2Cl$, pyridine, CH_2Cl_2, rt.

oxybenzaldehyde resin (DMHB resin), followed by alkylation and hydrolysis. Amide coupling of resin-bound acid **9** with diamine **6a** and subsequent resin cleavage provided compound **10a**, which was further elaborated into **10b–f** via reduction amination, acylation, or sulfonylation. Compounds **10g–r** were prepared in a similar manner as **10b** by replacing 3,4-dichloroaniline with various commercially available amines.

As shown in Table 4, the optimal R group is methyl (**10b**). While hydrogen (**10a**) and alkyl groups (**10b–d**) showed good binding affinity, acetyl (**10e**) had very poor affinity. Interestingly, methylsulfonyl (**10f**) was quite potent—indicating that the weakly basic center in this region was unnecessary for UT binding. 3,4-Dichlorophenyl (**10b**) was the optimal aryl (Ar) group. Bioisosteres such as 3,4-dimethylphenyl (**10g**) and 2-naphthyl (**10h**) were 10-fold less potent. *Meta*-substituted phenyl (**10i**) was preferred over *para*- (**10j**) and *ortho*- (**10k**) substituted ones. Comparing substituents on the phenyl ring, 3-chloro (**10i**) and 3-trifluoromethyl (**10m**) were preferred over 3-methyl (**10n**) and 3-methoxy (**10p**),

Table 4. SAR of the LHS moiety of the α-amino acetamide sub-series



Compound	Ar	R	hUT binding (pK_i)[a]
10a	3,4-Dichlorophenyl	Hydrogen	7.2
10b	3,4-Dichlorophenyl	Methyl	7.8
10c	3,4-Dichlorophenyl	Ethyl	7.5
10d	3,4-Dichlorophenyl	Cyclopropyl-methyl	7.3
10e	3,4-Dichlorophenyl	Acetyl	5.8
10f	3,4-Dichlorophenyl	Methylsulfonyl	7.1
10g	3,4-Dimethylphenyl	Methyl	6.7
10h	2-Naphthyl	Methyl	6.8
10i	3-Chlorophenyl	Methyl	7.1
10j	4-Chlorophenyl	Methyl	6.3
10k	2-Chlorophenyl	Methyl	6.1
10m	3-Trifluoromethyl-phenyl	Methyl	7.0
10n	3-Methylphenyl	Methyl	6.4
10p	3-Methoxyphenyl	Methyl	6.1
10q	Phenyl	Methyl	5.5
10r	3,4-Dichlorobenzyl	Methyl	5.4

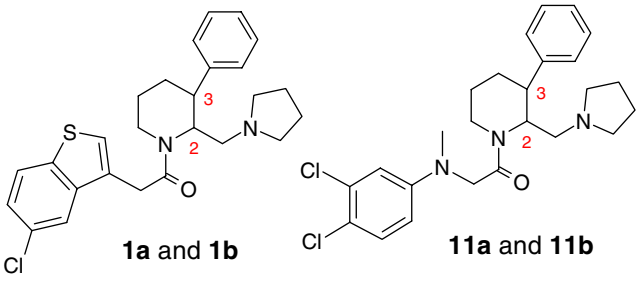
[a] Mean of at least 3 determinations with standard deviation of <±0.3.

and the unsubstituted phenyl (**10q**) was least preferred. Extending the phenyl to benzyl such as **10r** resulted in significant affinity loss.

We then investigated the preferred stereochemistry and found that (2*R*, 3*R*) was preferred as illustrated in Table 5. The (2*R*, 3*R*) enantiomers **1a** and **11a** were greater than 100-fold more potent than the (2*S*, 3*S*) enantiomers **1b** and **11b**.[17]

In addition to the high affinity to the hUT receptor, the preferred enantiomers **1a** and **11a** showed excellent

Table 5. Preferred stereochemistry



Compound	Stereochemistry and ee	hUT binding (pK_i)[a]
1a	2*R*, 3*R* (98% ee)	8.4
1b	2*S*, 3*S* (>99% ee)	6.0
11a	2*R*, 3*R* (98% ee)	8.2
11b	2*S*, 3*S* (>99% ee)	5.8

[a] Mean of at least 3 determinations with standard deviation of <±0.3.

Table 6. Binding affinity versus ortholog receptors and functional activity in rat and cat isolated arteries

Compound	UT binding (pK_i)[a]			Functional activity (pK_B)[a]	
	Human	Cat	Rat	Rat aorta	Cat femoral artery
1a	8.4	8.0	8.5	7.7	7.2
11a	8.2	8.1	8.2	7.9	7.2

[a] Mean of at least 3 determinations with standard deviation of <±0.3.

affinity to cat and rat UT receptors,[18] demonstrating broad cross-species activity (Table 6). In the rat isolated aorta,[18] compounds **1a** and **11a** blocked hU-II induced contraction with respective pK_Bs of 7.7 and 7.9, and **1a** was found to be a competitive and reversible antagonist with a pA_2 of 8.0, which was comparable to the rat receptor binding affinity. In the cat isolated femoral artery,[19] **1a** and **11a** also blocked hU-II induced contraction with pK_Bs of 7.2 and 7.2, respectively. The competitiveness and broad cross-species activity of **1a** distinguish it from early UT antagonists such as palosuran.[9c] However, several early compounds in the series had poor cytochrome P450 (CYP450) and in vivo pharmacokinetic (PK) parameters. For example, compound **10b** had 2D6 (IC_{50} 0.8 μM) and 3A4 (IC_{50} 1.4 μM) liability, and high clearance and low oral bioavailability in rat PK studies (CL 97 mL/min/kg, F 0–3%, $T_{1/2}$ 3.8 h, V_{dss} 23 L/kg, 1.2 mg/kg iv and 2.2 mg/kg po).

In summary, SAR exploration of a novel 2-aminomethyl piperidine series identified via HTS led to the discovery of competitive and reversible UT receptor antagonists such as **1a** and **11a** with excellent affinity and functional activity, and broad cross-species activity. The further optimization of this series to improve PK and CYP450 properties will be the subject of future publications.

Acknowledgments

We thank Doug Minick for VCD, Bing Wang for NMR, Qian Jin for LC/MS, Carl Bennett for purification, and Christina Schulz-Pritchard, Parvathi Rao, Jim Fornwald, Jeffrey Guss and Lorena Kallal for assay support.

References and notes

1. Ames, R. S.; Sarau, H. M.; Chambers, J. K.; Willette, R. N.; Aiyar, N. V.; Romanic, A. M.; Louden, C. S.; Foley, J. J.; Sauermelch, C. F.; Coatney, R. W.; Ao, Z.; Disa, J.; Holmes, S. D.; Stadel, J. M.; Martin, J. D.; Liu, W. S.; Glover, G. I.; Wilson, S.; McNulty, D. E.; Ellis, C. E.; Elshourbagy, N. A.; Shabon, U.; Trill, J. J.; Hay, D. W.; Ohlstein, E. H.; Bergsma, D. J.; Douglas, S. A. *Nature* **1999**, *401*, 282.
2. Douglas, S. A.; Dhanak, D.; Johns, D. G. *Trends Pharmacol. Sci.* **2004**, *25*, 76.
3. (a) Matsushita, M.; Shichiri, M.; Imai, T.; Iwashina, M.; Tanaka, H.; Takasu, N.; Hirata, Y. *J. Hypertens.* **2001**, *19*, 2185; (b) Cheung, B. M.; Leung, R.; Man, Y. B.; Wong, L. Y. *J. Hypertens.* **2004**, *22*, 1341.
4. Russell, F. D.; Meyers, D.; Galbraith, A. J.; Bett, N.; Toth, I.; Kearns, P.; Molenaar, P. *Am. J. Physiol.* **2003**, *285*, H1576.
5. (a) Douglas, S. A.; Tayara, L.; Ohlstein, E. H.; Halawa, N.; Giaid, A. *Lancet* **2002**, *359*, 1990; (b) Ng, L. L.; Loke, I.; O'Brien, R. J.; Squire, I. B.; Davies, J. E. *Circulation* **2002**, *106*, 2877; (c) Richards, A. M.; Nicholls, M. G.; Lainchbury, J. G.; Fisher, S.; Yandle, T. G. *Lancet* **2002**, *360*, 545; (d) Lapp, H.; Boerrigter, G.; Costello-Boerrigter, L. C.; Jaekel, K.; Scheffold, T.; Krakau, I.; Schramm, M.; Guelker, H.; Stasch, J. P. *Int. J. Cardiol.* **2004**, *94*, 93.
6. (a) Bousette, N.; Patel, L.; Douglas, S. A.; Ohlstein, E. H.; Giaid, A. *Atherosclerosis* **2004**, *176*, 117; (b) Maguire, J. J.; Kuc, R. E.; Wiley, K. E.; Kleinz, M. J.; Davenport, A. P. *Peptides* **2004**, *25*, 1767.
7. (a) Totsune, K.; Takahashi, K.; Arihara, Z.; Sone, M.; Satoh, F.; Ito, S.; Kimura, Y.; Sasano, H.; Murakami, O. *Lancet* **2001**, *358*, 810; (b) Shenouda, A.; Douglas, S. A.; Ohlstein, E. H.; Giaid, A. *J. Histochem. Cytochem.* **2002**, *50*, 885; (c) Langham, R. G.; Gow, R.; Thomson, N. M.; Dowling, J. K.; Gilbert, R. E. *Am. J. Kidney Dis.* **2004**, *44*, 826.
8. (a) Totsune, K.; Takahashi, K.; Arihara, Z.; Sone, M.; Satoh, F.; Ito, S.; Murakami, O. *Clin. Sci.* **2003**, *104*, 1; (b) Wenyi, Z.; Suzuki, S.; Hirai, M.; Hinokio, Y.; Tanizawa, Y.; Matsutani, A.; Satoh, J.; Oka, Y. *Diabetologi* **2003**, *46*, 972.
9. (a) Flohr, S.; Kurz, M.; Kostenis, E.; Brkovich, A.; Fournier, A.; Klabunde, T. *J. Med. Chem.* **2002**, *45*, 1799; (b) Croston, G. E.; Olsson, R.; Currier, E. A.; Burstein, E. S.; Weiner, D.; Nash, N.; Severance, D.; Allenmark, S. G.; Thunberg, L.; Ma, J.; Mohell, N.; O'Dowd, B.; Brann, M. R.; Hacksell, U. *J. Med. Chem.* **2002**, *45*, 4950; (c) Clozel, M.; Binkert, C.; Birker-Robaczewska, M.; Boukhadra, C.; Ding, S.; Fischli, W.; Hess, P.; Mathys, B.; Morrison, K.; Muller, C.; Muller, C.; Nayler, O.; Qiu, C.; Rey, M.; Scherz, M. W.; Velker, J.; Weller, T.; Xi, J.; Ziltener, P. *J. Pharma. Exp. Ther.* **2004**, *311*, 204; (d) Jin, J.; Dhanak, D.; Knight, S. D.; Widdowson, K.; Aiyar, N.; Naselsky, D.; Sarau, H. M.; Foley, J. J.; Schmidt, D. B.; Bennett, C. D.; Wang, B.; Warren, G. L.; Moore, M. L.; Keenan, R. M.; Rivero, R. A.; Douglas, S. A. *Bioorg. Med. Chem. Lett.* **2005**, *15*, 3229; (e) Douglas, S. A.; Behm, D. J.; Aiyar, N. V.; Naselsky, D.; Disa, J.; Brooks, D. P.; Ohlstein, E. H.; Gleason, J. G.; Sarau, H. M.; Foley, J. J.; Buckley, P. T.; Schmidt, D. B.; Wixted, W. E.; Widdowson, K.; Riley, G.; Jin, J.; Gallagher, T. F.; Schmidt, S. J.; Ridgers, L.; Christmann, L. T.; Keenan, R. M.; Knight, S. D.; Dhanak, D. *Br. J. Pharmacol.* **2005**, *145*, 620; (f) Lehmann, F.; Pettersen, A.; Currier, E. A.; Sherbukhin, V.; Olsson, R.; Hacksell, U.; Luthman, K. *J. Med. Chem.* **2006**, *49*, 2232; (g) Luci, D. K.; Ghosh, S.; Smith, C. E.; Qi, J.; Wang, Y.; Haertlein, B.; Parry, T. J.; Li, J.; Almond, H. R.; Minor, L. K.; Damiano, B. P.; Kinney, W. A.; Maryanoff, B. E.; Lawson, E. C. *Bioorg. Med. Chem. Lett.* **2007**, *17*, 6489; For a recent review, see: (h) Jin, J.; Douglas, S. A. *Expert Opin. Ther. Patents* **2006**, *16*, 467.
10. For radioligand binding and $[Ca^{2+}]_i$-mobilization assay details, see Ref. 9e.
11. The chiral centers in the structures shown in the paper are racemic except the ones in **1a**, **1b**, **11a** and **11b**.
12. The radioligand binding assay was used as the primary assay to generate SAR.
13. All new compounds in this paper were characterized via LC/MS and ^1H NMR.
14. Sakamoto, T.; Kaneda, S.; Nishimura, S.; Yamanaka, H. *Chem. Pharm. Bull.* **1985**, *3*, 565.
15. Diamine **6d** was produced as an over reduction by-product in the hydrogenation step from conversion of **5a** to **6a**.

16. Compounds **7e** and **7f** were prepared from the corresponding commercially available amino acids in the same way as the preparation of **3a** from **4** in Scheme 1.
17. (a) Intermediate **6a** was separated via chiral HPLC into two enantiomers, which were converted to **1a**, **1b**, **11a**, and **11b**. Chiral separation conditions: Chiralpak AD (77×240 mm, 20 μM) eluting at 300 mL/min with 0.1% isopropylamine in MeOH gave two components at R_T 3.3 and 5.1 min; (b) Stereochemistry was assigned by Vibrational Circular Dichroism (VCD) analysis of both enantiomers.
18. For cat and rat UT receptor radioligand binding and rat isolated aorta contractile assay details, see Ref. 9e.
19. For cat isolated femoral artery assay details, see: Behm, D. J.; Stankus, G.; Doe, C. P.; Willette, R. N.; Sarau, H. M.; Foley, J. J.; Schmidt, D. B.; Nuthulaganti, P.; Fornwald, J. A.; Ames, R. S.; Lambert, D. G.; Calo', G.; Camarda, V.; Aiyar, N. V.; Douglas, S. A. *Br. J. Pharmacol.* **2006**, *148*, 173.