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Synthesis and structure–activity relationships of biarylcarboxamide bis-aminopyrrolidine urea derived small-molecule antagonists of the melanin-concentrating hormone receptor-1 (MCH-R1)

Martin W. Rowbottom,[a,*] Troy D. Vickers,[a] Brian Dyck,[a] Junko Tamiya,[a] Mingzhu Zhang,[a] Liren Zhao,[a] Jonathan Grey,[a] David Provencal,[b] David Schwarz,[c] Christopher E. Heise,[d] Monica Mistry,[d] Andrew Fisher,[e] Teresa Dong,[e] Tao Hu,[a] John Saunders[a] and Val S. Goodfellow[a]

[a]*Department of Medicinal Chemistry, Neurocrine Biosciences Inc., 12790 El Camino Real, San Diego, CA 92130, USA*
[b]*Department of Process Chemistry, Neurocrine Biosciences Inc., 12790 El Camino Real, San Diego, CA 92130, USA*
[c]*Department of Molecular Biology, Neurocrine Biosciences Inc., 12790 El Camino Real, San Diego, CA 92130, USA*
[d]*Department of Pharmacology, Neurocrine Biosciences Inc., 12790 El Camino Real, San Diego, CA 92130, USA*
[e]*Department of Preclinical Development, Neurocrine Biosciences Inc., 12790 El Camino Real, San Diego, CA 92130, USA*

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Abstract—A novel series of bis-aminopyrrolidine ureas containing either a 4-biphenylcarboxmide or 5-phenyl-2-thiophenecarboxamide group have been identified as potent and functional antagonists of the melanin-concentrating hormone receptor-1. Syntheses and SAR are described, which led to the discovery of compounds with high binding affinity (K_i = 1 nM) for the receptor. Preliminary in vitro metabolic stability data are also reported for key compounds.
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Mammalian melanin-concentrating hormone (MCH) is a 19 amino acid cyclic peptide, which was first isolated from the brain tissue of rats.[1] Subsequently, it was discovered that in humans, MCH selectively binds and activates two G protein-coupled receptors, MCH-R1 and MCH-R2.[2] In the rat CNS, MCH peptide is strongly expressed within the lateral hypothalamus and the distribution of MCH-R1 strongly correlates to the expression of MCH peptide.[2,3] It is well known that the lateral hypothalamus plays a central role in the control of energy homeostasis, feeding behavior, and body weight. Indeed, there is mounting evidence suggesting that in rodents, MCH and the MCH-R1 receptor play a pivotal role in these processes. In rodents, increased brain levels of the MCH peptide lead to stimulation of food intake, obesity, and insulin resistance,[4] which is believed to occur through activation of the MCH-R1 receptor.[5] It has also been observed that mice lacking MCH are both hypophagic and lean.[4] Interestingly, MCH-R1 deficient mice (mchr1−/−) are lean, hyperphagic, have altered metabolism, and are resistant to diet-induced obesity, suggesting that MCH and the MCH-R1 receptor have a role in controlling metabolic processes as well as regulation of appetite.[4] Therefore, the development of selective MCH-R1 antagonists may be useful for the treatment of obesity and related diseases. Both peptide[6] and small-molecule antagonists[7] of the MCH-R1 receptor have been reported to suppress MCH-stimulated food intake in rodents. In addition, recent studies suggest that MCH may be involved in the regulation of stress behavior,[4,8] and it was discovered that potent and selective small-molecule antagonists of the MCH-R1 receptor displayed anxiolytic and antidepressant properties in animal models.[7b,8b] It is therefore not surprising that there is currently great interest in the development of small-molecule MCH-R1 receptor

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* Corresponding author. Tel.: + 1 858 617 7799; fax: +1 858 617 7601; e-mail: mrowbottom@neurocrine.com
URL: http://www.neurocrine.com

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antagonists, which hold the potential to treat both obesity and stress related disorders.[7,8b,9]

As part of our efforts to develop novel medicines focused on the treatment of CNS disorders, we initiated research toward discovery of orally active small-molecule MCH-R1 antagonists. Previously, we discovered that bis-aminopyrrolidine urea **1** (Fig. 1), containing both an *N*-3-(4-chlorophenyl)propyl chain and a 4-(4-trifluoromethyl)biphenyl substituted methylcarboxamide, was a potent and functional antagonist of the MCH-R1 receptor.[9i] Unfortunately, the in vitro metabolic stability (in human liver microsomes) of bis-aminopyrrolidine urea **1** proved to be low, suggesting that it would be rapidly cleared in vivo. This may be partly due to the molecule having a relatively high log *P* (for compound **1**, the calculated log *P* is 6.4).[10] Removal of the large 3-(4-chlorophenyl)propyl side chain would result in analogs with a reduced log *P*, in the range of 2–3 log units. We therefore explored a series of bis-aminopyrrolidine ureas containing an *N*-methyl substituent (represented by **2**, Fig. 1). These compounds proved to be potent antagonists of the MCH-R1 receptor with improved drug-like properties. In this letter, we wish to report the synthesis and SAR of this novel class of small-molecule MCH-R1 antagonists. In particular, changes around the biarylcarboxamide portion of the molecule will be described and the resulting SAR will be discussed, along with preliminary metabolic stability data for this series.

All biarylcarboxamide bis-aminopyrrolidine urea derivatives **2** were synthesized from the Boc-protected core **6**, which was prepared as outlined in Scheme 1. Reaction of commercially available (*R*)-(−)-1-benzyl-3-(methylamino)pyrrolidine with 4-nitrophenylchloroformate gave (*R*)-1-benzyl-3-[(4-nitrophenoxycarbonyl)methylamino]-pyrrolidinium hydrochloride **3** in 83% yield. Reaction of pyrrolidinium salt **3** with commercially available (*R*)-(−)-3-pyrrolidinol hydrochloride in the presence of TEA gave pyrrolidinol urea **4** in 77% yield. The hydroxyl group of **4** was activated using 4-nitrophenylsulfonyl chloride to give the nosylate intermediate, which was then displaced by methylamine, followed by reaction with di-*tert*-butyl-dicarbonate to give N-Boc protected (*S*)-3-(methylamino)pyrrolidine-1-carboxylic acid [(*R*)-1-benzylpyrrolidin-3-yl]methylamide **5** in 68% yield (from **4**), as a single diastereoisomer.[11] N-Debenzylation, followed by reductive alkylation with formaldehyde, gave N-Boc protected (*S*)-3-[(*N*)-methylamino] pyrrolidine-1-carboxylic acid [(*R*)-1-methylpyrrolidin-3-yl]methylamide **6** in 89% yield. N-Boc deprotection (of **6**), followed by coupling with either 4-iodobenzoic acid or 5-bromothiophene-2-carboxylic acid, gave the key intermediates 4-iodophenylcarboxamide **7** (in 83% yield) and 5-bromo-2-thiophenecarboxamide **8** (in 77% yield), respectively. The reaction of 4-iodophenylcarboxamide **7** or 5-bromo-2-thiophenecarboxamide **8** with a variety of substituted phenylboronic acids (via a Suzuki coupling) gave the desired 4-biphenylcarboxamides **9**–**11** and 5-phenyl-2-thiophenecarboxamides **12**–**42**, respectively (Scheme 2 and Tables 1–3).[12] All biarylcarboxamide bis-aminopyrrolidine ureas described were assayed for their ability to displace radiolabeled [^{125}I-Tyr^{13}]MCH in a competitive binding assay.[13] The assay was performed using a human MCH-R1 receptor that is modified for optimal expression in HEK293 cells. Selected results are summarized in Tables 1–3. The functional antagonism of all compounds with measured K_i's less than 50 nM was further confirmed based on their ability to inhibit, in a dose-dependent manner, MCH stimulated G protein–[^{35}S]–GTPγS binding in cells expressing the native human MCH-R1 receptor (as represented by compound **35**, Fig. 2).[14] In addition, the compounds described were shown to be inactive against human MCH-R2[15] and optimized compounds exhibited good selectivity when assayed against a standard panel of GPCRs, including those of the serotonin, dopamine, opioid, and muscarinic family of receptors.[16]

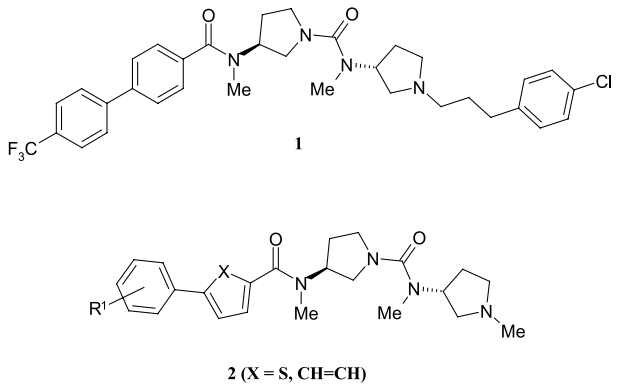
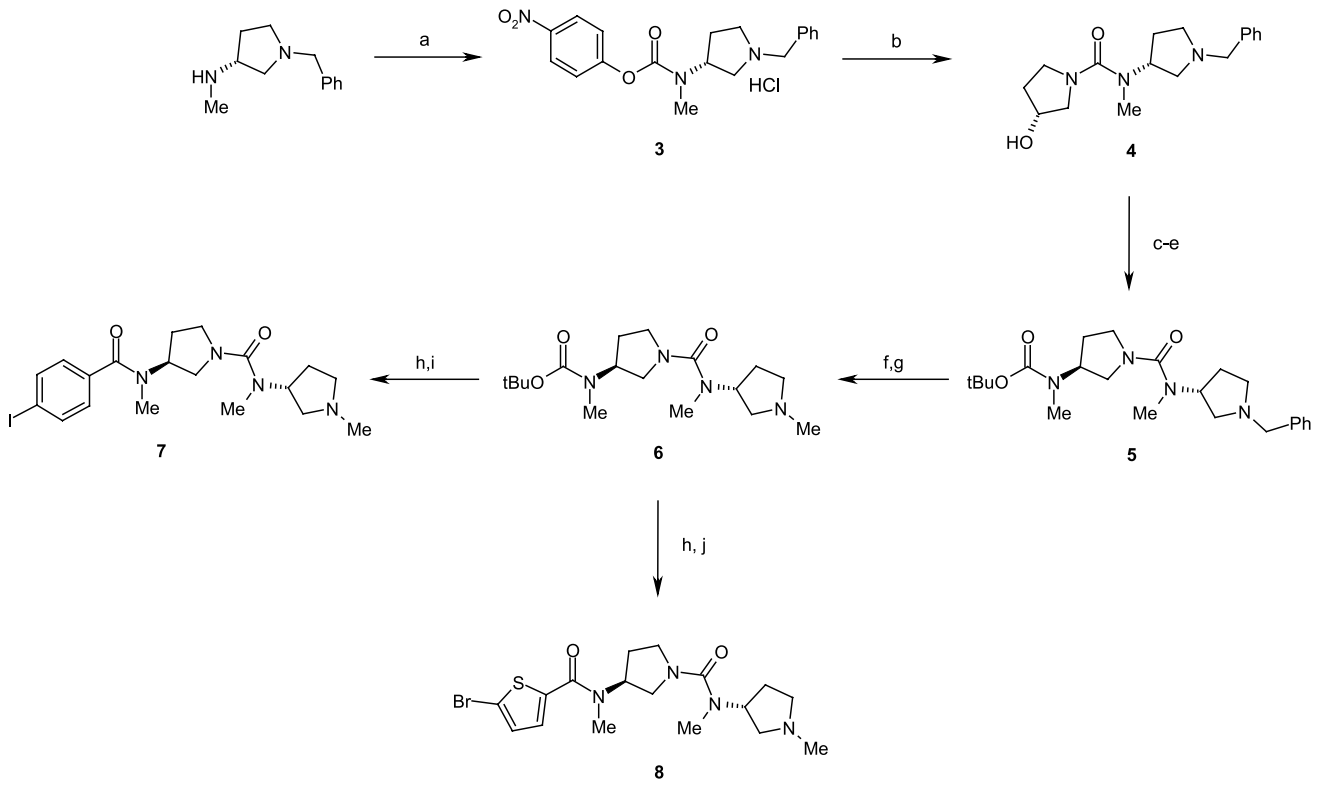
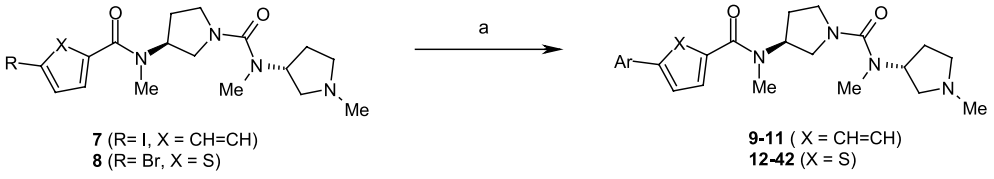


Figure 1. General structures of bis-aminopyrrolidine urea MCH-R1 antagonists.

Bis-aminopyrrolidine ureas containing a 4-biphenylcarboxamide group (see compounds **9**–**11**, Table 1) are very potent antagonists of the MCH-R1 receptor. It was observed that the terminal phenyl ring could either be electron rich or electron deficient (see compounds **9**–**11**). Not surprisingly, when the central phenyl ring was replaced with a thiophene ring, potency was maintained (see compounds **12**–**15**, Table 1). Incorporation of a methyl substituent *ortho* to the biaryl bond did not significantly affect potency in the case of the 5-phenyl-2-thiophenecarboxamide derivatives (compare compounds **14** and **15**), but potency was slightly decreased in the case of the 4-biphenyl derivatives. The 4-(4-methoxy-2-methylphenyl)phenylcarboxamide derivative **11** had a K_i of 16 nM, which was approximately 3-fold less potent than the corresponding des-methyl analog **10** (K_i = 5 nM). We also explored the effect of changing the absolute configuration of the two stereocenters, as summarized in Table 2. The three other diastereoisomers of 5-[4-(trifluoromethyl)phenyl]-2-thiophenecarboxamide derivative **12** were prepared (see compounds **16**–**18**, Table 2).[17] The (*S*, *R*) diasteroisomer **12** proved to be the most potent with a measured K_i of 2 nM. The (*S*, *S*) diasteroisomer **16** was only 7-fold less potent (K_i = 14 nM), whereas both the (*R*, *S*) (compound **17**) and (*R*, *R*) (compound **18**) diasteroisomers were at least 75-fold less active.



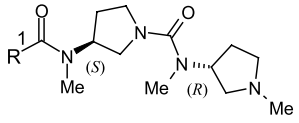
Scheme 1. Reagents and conditions: (a) 4-nitrophenyl chloroformate, THF, 5 °C to rt, 3 h, 83%; (b) (*R*)-(−)-3-pyrrolidinol hydrochloride, TEA, DMF, 70 °C, 18 h, 77%; (c) 4-nitrobenzenesulfonyl chloride, TEA, THF, 5 °C to rt, 18 h; (d) methylamine, THF/H_2O, 50 °C, 18 h; (e) $(Boc)_2O$, THF, rt, 30 min, 68% (from **4**); (f) H_2 (40 psi), $Pd(OH)_2$/C, EtOH, rt, 17 h, quantitative; (g) formaldehyde, $NaBH(OAc)_3$, 1,2-DCE/H_2O, rt, 4 h, 89%; (h) TFA, DCM, rt, 1 h, quantitative; (i) 4-iodobenzoic acid, EDC, HOBT, TEA, DCM, rt, 20 h, 83%; (j) 5-bromothiophene-2-carboxylic acid, EDC, HOBT, TEA, DCM, rt, 20 h, 77%.

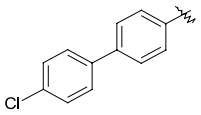
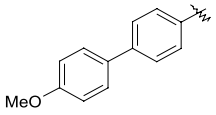
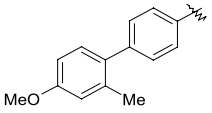
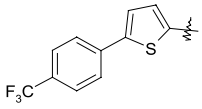
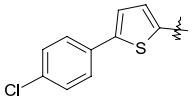
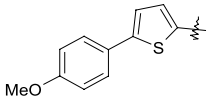
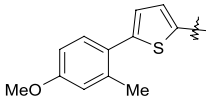


Scheme 2. Reagents and conditions: (a) $ArB(OH)_2$, $Pd(PPh_3)_4$, Na_2CO_3, 1,4-dioxane/H_2O, reflux, 12 h.

Using 5-[4-(trifluoromethyl)phenyl]-2-thiophenecarboxamide bis-aminopyrrolidine urea **12** as a lead structure, substitution around the terminal phenyl ring was explored and selected results are summarized in Table 3. Moving the trifluoromethyl group to the 3-position of the phenyl ring led to a dramatic loss in potency. The 3-(trifluoromethyl)phenyl derivative **19** had a K_i of 1,800 nM and was approximately 900-fold less potent than the 4-(trifluoromethyl)phenyl analog **12** (K_i = 2 nM; see Table 1). The 2-(trifluoromethyl)phenyl derivative **20** (K_i of 61 nM) also proved to be less active than the 4-substituted analog **12**. It was generally observed that compounds containing a *meta*-substituted phenyl ring were not well tolerated by the receptor. For instance, both the 3-chlorophenyl **21** (K_i = 320 nM) and 3-methoxyphenyl **23** (K_i = 900 nM) derivatives were much less active than their corresponding 4-substituted analogs **13** (K_i = 2 nM) and **14** (K_i = 4 nM), respectively (see Table 1). Interestingly, the 2-chlorophenyl derivative **22** had a K_i of 10 nM, which proved to be 6-fold more potent than the 2-(trifluoromethyl)phenyl derivative **20** and over 300-fold more potent than the corresponding 2-methoxyphenyl analog **24** (K_i = 3,400 nM). In addition, the 2-methylphenyl derivative **26** had a K_i of 19 nM, whereas the 2,6-dimethylphenyl derivative **28** was 16-fold less potent (K_i of 310 nM). This shows that compounds containing a 2,6-disubstituted phenyl are less well tolerated by the receptor. This may be due to a certain steric interaction or that the phenyl ring is forced to adopt an unfavorable conformation (relative to the adjacent thiophene). Importantly, the 5-(2,4-dimethylphenyl)-2-thiophenecarboxamide derivative **27** had a K_i of 3 nM, this being 6-fold more potent than the 2-methylphenyl analog **26**. Together these results emphasize the importance of *para*-substitution (on the phenyl ring) for optimal receptor binding and a more detailed study to fully explore the SAR of this key substituent was carried out.

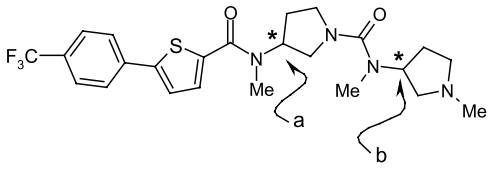
Table 1. Binding affinities of bis-aminopyrrolidine ureas **9**–**15** toward the MCH-R1 receptor



Compound	R^1	K_i (nM)[a]
9		2
10		5
11		16
12		2
13		2
14		4
15		3

[a] Data are average of three or more independent measurements.[13]

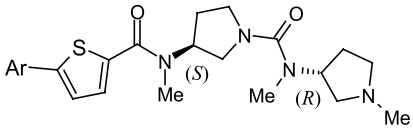
Table 2. Binding affinities of bis-aminopyrrolidine ureas **12**, **16**–**18** toward the MCH-R1 receptor

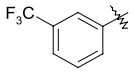
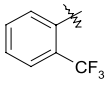
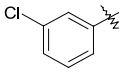
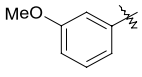
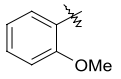
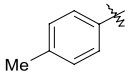
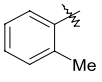
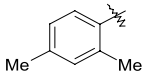
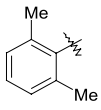
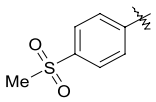


Compound	a,b	K_i (nM)[a]	Optical purity (%)[b]
12	*S*,*R*	2	>97
16	*S*,*S*	14	>97
17	*R*,*S*	190	>97
18	*R*,*R*	150	>97

[a] Data are average of three or more independent measurements.[13]
[b] Experimentally determined.[17]

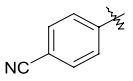
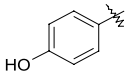
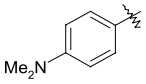
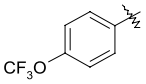
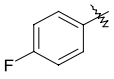
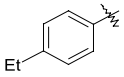
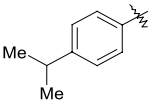
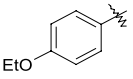
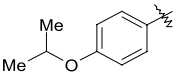
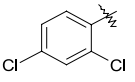
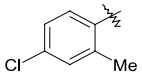
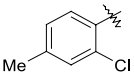
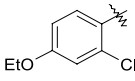
Table 3. Binding affinities of bis-aminopyrrolidine ureas **19**–**42** toward the MCH-R1 receptor



Compound	Ar	K_i (nM)[a]
19		1,800
20		61
21		320
22		10
23		900
24		3,400
25		3
26		19
27		3
28		310
29		6,000

(*continued on next page*)

Table 3 (*continued*)

Compound	Ar	K_i (nM)[a]
30		2,400
31		>10,000
32		7
33		4
34		37
35		**1**
36		5
37		63
38		3,300
39		3
40		3
41		**1**
42		2

[a] Data are average of three or more independent measurements.[13]

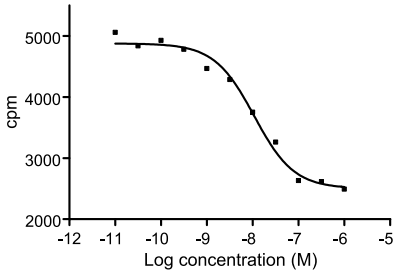


Figure 2. Inhibition of MCH (10 nM) stimulated G protein–[^{35}S]–GTPγS binding, by compound **35**. Compound **35** has a measured IC_{50} of 14 nM (n = 3).[14]

Incorporation of relatively polar or bulky functionality at the 4-position of the phenyl ring had a detrimental effect on potency, though the electronic nature of this substituent was not important, as compounds containing either an electron rich or electron deficient phenyl ring were well tolerated by the receptor. The 4-(methylsulfonyl)phenyl **29**, 4-cyanophenyl **30**, and 4-hydroxyphenyl **31** derivatives (Table 3) had K_i's of 6000, 2400, and >10,000 nM, respectively, these substituents being too polar in nature. Derivatives incorporating more lipophilic substituents proved to be much more potent. For instance, compounds **32**, **33**, and **35** had K_i's of 7, 4, and 1 nM, respectively, a level of activity not witnessed with the slightly less lipophilic 4-fluorophenyl analog **34**. Indeed the 4-ethylphenyl derivative **35** proved to be one of the most potent analogs prepared in this series. With the introduction of the slightly larger isopropyl group (compound **36**, K_i = 5 nM) potency was maintained. However, introduction of progressively larger groups led to a noticeable reduction in potency (compare compound **14** with both compounds **37** and **38**). Potency may be improved by addition of a suitable substituent *ortho* to the biaryl bond. For instance, the 2-chloro-4-ethoxyphenyl derivative **42** had a K_i of 2 nM and was approximately 30-fold more potent than the 4-ethoxyphenyl analog **37** (K_i = 63 nM). This suggests that the 2-chloro substituent may help to orient the phenyl ring into a particular conformation (relative to the adjacent thiophene), which may be preferred for optimal receptor binding. In addition, the 2,4-dichlorophenyl **39**, 4-chloro-2-methylphenyl **40**, and 2-chloro-4-methylphenyl **41** derivatives had K_i's of 3, 3, and 1 nM, respectively. Together these data suggest that the receptor binding pocket in which the phenyl (of the 5-phenyl-2-thiophenecarboxamide moiety) sits is relatively tight and lipophilic in nature. A 4-substituted or 2,4-disubstituted phenyl is preferred, whereas 3-substituents are not well tolerated. In addition, the presence of either a relatively polar or bulky substituent (at the 2- or 4-position) tends to reduce binding affinity with the receptor.

Ultimately our focus was on the discovery of orally active small-molecule antagonists of the MCH-R1 receptor, so we were particularly interested in the PK profile of this novel series of compounds. Incubation with human liver microsomes (HLM) can be used to estimate how much of a target compound will be re-

moved by hepatic first-pass metabolism in vivo. For instance in an in vitro HLM assay,[18] 5-(4-ethylphenyl)-2-thiophenecarboxamide derivative **35** (Table 3) had an intrinsic clearance of 95 mL/min/kg, implying that this compound is being rapidly metabolized. Other examples proved to be much more stable to metabolism by HLM. For instance, the 2-chloro-4-methylphenyl **41** and 2-chloro-4-ethoxyphenyl **42** analogs (Table 3) had intrinsic clearance values of 19 and 6 mL/min/kg, respectively. Inhibition of certain cytochrome P450 (CYP450) metabolizing enzymes (particularly CYP2D6 and CYP3A4) may lead to undesirable drug–drug interactions in vivo. The compounds described proved to be very weak inhibitors of both CYP2D6 and CYP3A4. For instance, in vitro the aforementioned compounds **35**, **41**, and **42** all had measured IC_{50} values >20 μM, against both CYP2D6 and 3A4.[19]

In conclusion, we have discovered a novel series of functional MCH-R1 antagonists based on an N-methylated bis-aminopyrrolidine urea core, containing either a 4-biphenylcarboxamide or 5-phenyl-2-thiophenecarboxamide group. SAR around the 5-phenyl-2-thiophenecarboxamide group was explored, where it was observed that either a 4-substituted or 2,4-disubstituted phenyl ring was preferred for optimal binding with the receptor. In addition, examples from this series display promising in vitro metabolic stability profiles, exhibiting good stability in the HLM assay with no significant inhibition of key CYP450 metabolizing enzymes. This new series proved to be significantly more stable (in vitro) than previously described first-generation bis-aminopyrrolidine urea MCH-R1 antagonists. Further results, including in vivo PK and efficacy data in animal models, will be reported in due course.

Acknowledgments

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References and notes

1. Vaughan, J. M.; Fischer, W. H.; Hoeger, C.; Rivier, J.; Vale, W. *Endocrinology* **1989**, *125*, 1660.
2. Boutin, J. A.; Suply, T.; Audinot, V.; Rodriguez, M.; Beauverger, P.; Nicolas, J.-P.; Galizzi, J.-P.; Fauchère, J.-L. *Can. J. Physiol. Pharmacol.* **2002**, *80*, 388, and references cited therein.
3. For binding sites within the human brain, see, Sone, M.; Takahashi, K.; Murakami, O.; Totsune, K.; Arihara, Z.; Satoh, F.; Sasano, H.; Ito, H.; Mouri, T. *Peptides* **2000**, *21*, 245.
4. Hervieu, G. *Expert Opin. Ther. Targets* **2003**, *7*, 495, and references cited therein.
5. Suply, T.; Della-Zuana, O.; Audinot, V.; Rodriguez, M.; Beauverger, P.; Duhault, J.; Canet, E.; Galizzi, J.-P.; Nahon, J.-L.; Levens, N.; Boutin, J. A. *J. Pharmacol. Exp. Ther.* **2001**, *299*, 137.
6. Shearman, L. P.; Camacho, R. E.; Stribling, D. S.; Zhou, D.; Bednarek, M. A.; Hreniuk, D. L.; Feighner, S. D.; Tan, C. P.; Howard, A. D.; Van der Ploeg, L. H. T.; MacIntyre, D. E.; Hickey, G. J.; Strack, A. M. *Eur. J. Pharmacol.* **2003**, *475*, 37.
7. (a) Takekawa, S.; Asami, A.; Ishihara, Y.; Terauchi, J.; Kato, K.; Shimomura, Y.; Mori, M.; Murakoshi, H.; Kato, K.; Suzuki, N.; Nishimura, O.; Fujino, M. *Eur. J. Pharmacol.* **2002**, *438*, 129; (b) Borowsky, B.; Durkin, M. M.; Ogozalek, K.; Marzabadi, M. R.; DeLeon, J.; Heurich, R.; Lichtblau, H.; Shaposhnik, Z.; Daniewska, I.; Blackburn, T. P.; Branchek, T. A.; Gerald, C.; Vaysse, P. J.; Forray, C. *Nat. Med.* **2002**, *8*, 825.
8. (a) Kela, J.; Salmi, P.; Rimondini-Giorgini, R.; Heilig, M.; Wahlestedt, C. *Regul. Pept.* **2003**, *114*, 109; (b) Kennedy, A. R.; Todd, J. F.; Dhillo, W. S.; Seal, L. J.; Ghatei, M. A.; O'Toole, C. P.; Jones, M.; Witty, D.; Winborne, K.; Riley, G.; Hervieu, G.; Wilson, S.; Bloom, S. R. *J. Neuroendocrinol.* **2003**, *15*, 268.
9. (a) Carpenter, A. J.; Hertzog, D. L. *Expert Opin. Ther. Patents* **2002**, *12*, 1639; (b) Collins, C. A.; Kym, P. R. *Curr. Opin. Investig. Drugs* **2003**, *4*, 386; (c) Clark, D. E.; Higgs, C.; Wren, S. P.; Dyke, H. J.; Wong, M.; Norman, D.; Lockey, P. M.; Roach, A. G. *J. Med. Chem.* **2004**, *47*, 3962; (d) Arienzo, R.; Clark, D. E.; Cramp, S.; Daly, S.; Dyke, H. J.; Lockey, P.; Norman, D.; Roach, A. G.; Stuttle, K.; Tomlinson, M.; Wong, M.; Wren, S. P. *Bioorg. Med. Chem. Lett.* **2004**, *14*, 4099; (e) Souers, A. J.; Wodka, D.; Gao, J.; Lewis, J. C.; Vasudevan, A.; Gentles, R.; Brodjian, S.; Dayton, B.; Ogiela, C. A.; Fry, D.; Hernandez, L. E.; Marsh, K. C.; Collins, C. A.; Kym, P. R. *Bioorg. Med. Chem. Lett* **2004**, *14*, 4873; (f) Vasudevan, A.; Wodka, D.; Verzal, M. K.; Souers, A. J.; Gao, J.; Brodjian, S.; Fry, D.; Dayton, B.; Marsh, K. C.; Hernandez, L. E.; Ogiela, C. A.; Collins, C. A.; Kym, P. R. *Bioorg. Med. Chem. Lett.* **2004**, *14*, 4879; (g) Souers, A. J.; Wodka, D.; Gao, J.; Lewis, J. C.; Vasudevan, A.; Gentles, R.; Brodjian, S.; Dayton, B.; Ogiela, C. A.; Fry, D.; Hernandez, L. E.; Marsh, K. C.; Collins, C. A.; Kym, P. R. *Bioorg. Med. Chem. Lett.* **2004**, *14*, 4883; (h) Receveur, J.-M.; Bjurling, E.; Ulven, T.; Little, P. B.; Nørregaard, P. K.; Högberg, T. *Bioorg. Med. Chem. Lett.* **2004**, *14*, 5075; (i) Grey, J.; Dyck, B.; Rowbottom, M. W.; Tamiya, J.; Vickers, T. D.; Zhang, M.; Zhao, L.; Heise, C. E.; Schwarz, D.; Saunders, J.; Goodfellow, V. S. *Bioorg. Med. Chem. Lett.* **2005**, *15*, 999; (j) Su, J.; McKittrick, B. A.; Tang, H.; Czarniecki, M.; Greenlee, W. J.; Hawes, B. E.; O'Neill, K. *Bioorg. Med. Chem.* **2005**, *13*, 1829.
10. Calculated using ACD/Labs Log P database, version 6.0 (2002), Advanced Chemistry Development Inc., Toronto, Ontario, Canada (http://www.acdlabs.com).
11. The diastereomeric purity of compound **5** was >98%, as measured by HPLC. This was determined by direct comparison to an equal mixture of both possible diastereomeric products, *N*-Boc protected (*R*)- and (*S*)-3-[(*N*-)methylamino]pyrrolidine-1-carboxylic acid [(*R*)-1-benzylpyrrolidin-3-yl]methylamide, which was prepared as described (Scheme 1) using commercially available racemic 3-pyrrolidinol hydrochloride
12. Analytical data for a typical example, the hydrochloride salt of 5-(4-trifluoromethylphenyl)-2-thiophenecarboxamide bis-aminopyrrolidine urea **12**: ^{1}H NMR δ_H (300 MHz; $CDCl_3$) 12.83 and 12.40 (2× br m, 1H), 7.73 (d, J = 8.3 Hz, 2H), 7.66 (d, J = 8.3 Hz, 2H), 7.38 (m, 1H), 7.34 (d, J = 3.3 Hz, 1H), 5.04 (m, 1H), 4.76 and 4.17 (2× m, 1H), 3.82 (m, 1H), 3.41–3.63 (m, 6H), 3.27 (m, 1H), 3.19 (s, 3H), 2.74–2.98 (m, 7H), 2.52 (m, 1H), 2.12–2.33 (m, 2H); MS (CI) *m/z*: 495.0 (M+H); Anal. Calcd for

$C_{24}H_{29}F_3N_4O_2S$–HCl–H_2O: C, 52.50; H, 5.87; N, 10.20; S, 5.84. Found: C, 52.39; H, 6.22; N, 10.26; S, 6.23.

13. On each assay plate, a standard antagonist of comparable affinity to those being tested was included as a control for plate-to-plate variability. Overall K_i values were highly reproducible with an average standard deviation of <45% for replicate determinations. All compounds described were assayed in at least three independent experiments.
14. Assays were performed using membrane preparations of CHO cells stably expressing the human MCH-R1 receptor. Each experiment was run in the presence of MCH peptide (10 nM), [γ-^{35}S]GTP (0.5 nM), and GDP (10 μM). On each assay plate, a standard antagonist of comparable IC_{50} to those being tested was included as a control for plate-to-plate variability. Overall IC_{50} values were highly reproducible with an average standard deviation of <45% for replicate determinations. All key compounds (K_i < 50 nM) were assayed in at least three independent experiments. In addition, representative compounds from this series behaved as functional antagonists in a FLIPR based Ca^{2+} flux assay, using cells stably expressing the hMCH-R1 receptor.
15. For example, in an hMCH-R2 binding assay compound **12** had a measured IC_{50} > 100 μM and was inactive in a corresponding Ca^{2+} flux functional assay.
16. Cross-reactivity assays were performed by CEREP (www.cerep.com). For example, compound **12** displayed >500-fold selectivity when assayed against more than 75 GPCR receptors.
17. Each diastereoisomer was prepared from commercially available optically pure building blocks. Measured by reverse-phase chiral HPLC (at 254 nm), the optical purity of each was determined to be >97% (when directly compared to a mixture of all four diastereoisomers). Analytical conditions: Chiralpak®AD™-RH column, dimensions 0.46 × 15 cm. Flow rate of 0.5 mL/min, isocratic elution with acetonitrile/water (9:1) (containing 0.1% diethylamine).
18. General experimental details for this assay may be found in the following reference. Guo, Z.; Zhu, Y.-F.; Gross, T. D.; Tucci, F. C.; Gao, Y.; Moorjani, M.; Connors, P. J.; Rowbottom, M. W.; Chen, Y.; Struthers, R. S.; Xie, Q.; Saunders, J.; Reinhart, G.; Chen, T. K.; Bonneville, A. L. K.; Chen, C. *J. Med. Chem.* **2004**, *47*, 1259.
19. Inhibition assays were carried out using microsomes isolated from transfected cells expressing only the relevant CYP450 isoform (2D6 or 3A4) and in the presence of a fluorescent substrate (AMMC for 2D6 and BFC for 3A4). Quinidine and ketoconazole were used as positive controls for the CYP2D6 and 3A4 assays, respectively.