Bioorganic & Medicinal Chemistry Letters 23 (2013) 4141–4144

Contents lists available at SciVerse ScienceDirect

Bioorganic & Medicinal Chemistry Letters

journal homepage: www.elsevier.com/locate/bmcl

The discovery of potent selective NPY Y_2 antagonists

Wenying Chai *, Victoria D. Wong, Diane Nepomuceno, Pascal Bonaventure, Timothy W. Lovenberg, Nicholas I. Carruthers

Janssen Research & Development LLC, 3210 Merryfield Row, San Diego, CA 92121, USA

ARTICLE INFO

Article history:
Received 20 February 2013
Revised 6 May 2013
Accepted 13 May 2013
Available online 22 May 2013

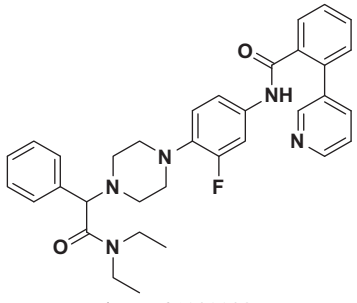
Keywords:
NPY Y_2
NPY Y_2 antagonists

ABSTRACT

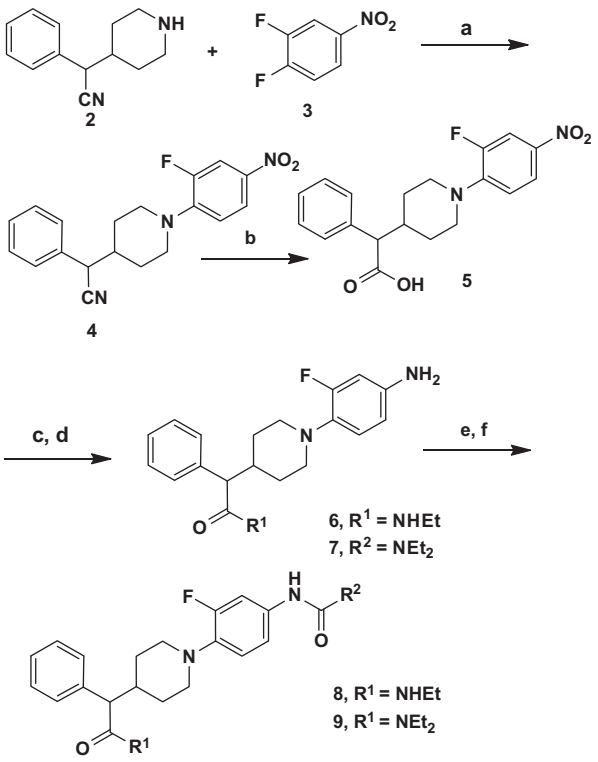
A series of small molecules with a piperidinyl core were synthesized and tested for binding affinity (IC_{50}) at human Neuropeptide Y Y_2 receptor. Various amide related analogs (ureas, reversed amides, and sulfonamides) were evaluated. Several potent and selective NPY Y_2 antagonists were identified.

Neuropeptide Y (NPY) is 36-amino acid peptide discovered in the early 1980s. It is widely distributed in the central and peripheral nervous systems.[1] To date, five NPY receptors (Y_1, Y_2, Y_4, Y_5, y_6) were identified. The Y_2 receptor is the predominant NPY receptor in the brain which can also be found in the periphery. The Y_2 receptor has been implicated in many physiological functions including bone formation, gastrointestinal motility, and food intake.[2,3] Identification of novel Y_2 receptor ligands will allow for further elucidation of the physiological role of the Y_2 receptor.[4–7]

NPY is the natural NPY Y_2 agonist. Several small molecule antagonists have been reported in the literature in recent years.[8–15] Some of them were described from our research lab.[14,15]



1, JNJ 31020028
NPY Y_2 IC_{50} = 6 nM
MTP IC_{50} = 710 nM

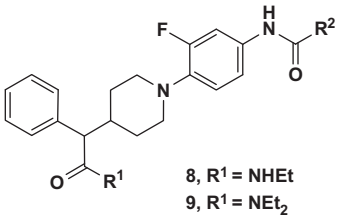


Scheme 1. Reagents and conditions: (a) K_2CO_3, DMF, 16 h, 95%; (b) 48% HBr, reflux, 90%; (c) $(COCl)_2$, DMF, CH_2Cl_2; (d) NH_2Et or $NHEt_2$, NEt_3, CH_2Cl_2; (c, d) 75–81%; (e) $SnCl_2{\cdot}2H_2O$, EtOH/EtOAc; (f) R^2 COCl, NEt_3, CH_2Cl_2; e to f: 38-72%.

* Corresponding author. Tel.: +1 8587843047.
E-mail address: wchai@its.jnj.com (W. Chai).

0960-894X/$ - see front matter © 2013 Elsevier Ltd. All rights reserved.
http://dx.doi.org/10.1016/j.bmcl.2013.05.038

Table 1
Amides



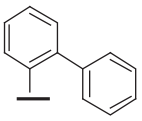
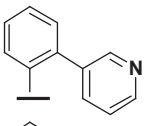
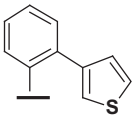
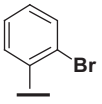
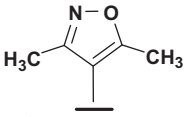
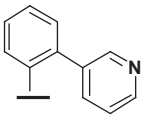
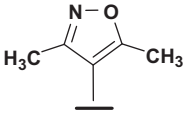
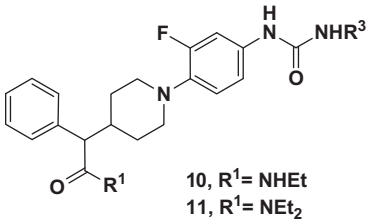
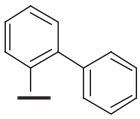
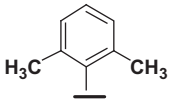
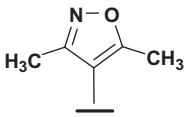
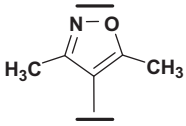
No.	R^2	NPY Y_2 IC_{50} (nM)[18]
8a		12 ± 4
8b		24 ± 11
8c		70 ± 50
8d		665 ± 135
8e		476 ± 116
9a		7 ± 1
9b		37 ± 23

Table 2
Ureas



No.	R^3	NPY Y_2 IC_{50} (nM)[18]
10a		27 ± 7
10b		162 ± 18
10c		15 ± 2
11a		12 ± 6

In order to identify selective, brain-penetrant, non-peptidic NPY Y_2 antagonists, a pharmacophore directed screen of our compound collection was initiated. Compound **1** was identified and reported in 2010.[14,15] The original lead from HTS was prepared for a microsomal triglyceride transfer protein (MTP) program.[16,17] Inhibiting MTP prevent the assembly of apo B-containing lipoproteins thus inhibiting the synthesis of chylomicrons and VLDL and leading to decrease in plasma levels of LDL-C. Thus, obtaining good selectivity over MTP was one of the goals for this project. Here we report a series of compounds related to **1**, with a piperidine core, as NPY Y_2 antagonists. Throughout the study, we identified potent NPY Y_2 compounds maintaining good MTP selectivity, and/or improved microsomal stability. We also identified peripherally restricted compounds. SAR and preliminary in vivo data for selected compounds are presented.

Our efforts focused on the right hand side biaryl amide portion. Ureas, reversed amide and sulfonamide analogs were evaluated. These compounds were synthesized and tested for NPY Y_2 binding affinity at the human receptor.[18] Chemistry started from 2-phenyl-2-(piperidin-4-yl)acetonitrile **2** reacting with 3,4-difluoro-nitrobenzne **3**, and K_2CO_3 in DMF. Amino replacement with fluorine was successful providing **4** in very good yield (95%). Hydrolysis of the cyano group with 48% HBr provided the corresponding carboxylic acid **5** (90%). Acid chloride formation followed by treatment with ethyl amine or diethyl amine yielded amides **6** and **7** (75–81%). Nitro reduction of **6** or **7** with $SnCl_2{\cdot}2H_2O$ in EtOH/EtOAc followed by amide formation with various acid chlorides providing the diamides **8** and **9** (38–72%) (Scheme 1).

The di-amides (Table 1) with biaryl groups at the right hand side **8a–8c** show good human binding affinity at NPY Y_2 (IC_{50}: 12–70 nM). The bromo intermediate **8d** also showed some affinity (IC_{50}: 665 nM). 3,5-Dimethyl oxazole **8e** was evaluated and showed affinity at NPY Y_2 (IC_{50}: 476 nM). Compounds with diethyl amides at the left hand side (**9a**, **9b**) show much better binding affinity (IC_{50}: 7–37 nM) compared to their mono-ethyl amide analogs (**8b**, **8d**).

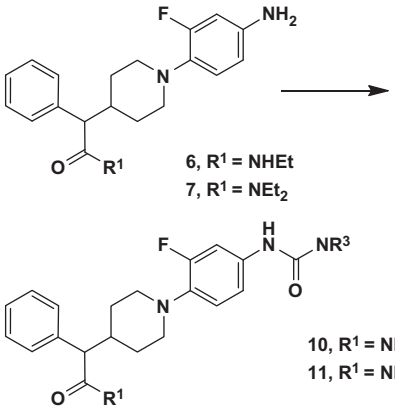
On the right hand side, urea analogs (Table 2) were prepared. Ureas were obtained through reacting **6** or **7** with different isocyanates (Scheme 2).

The urea with a biphenyl group at the right hand side (**10a**) demonstrated good binding affinity at NPY Y_2 (IC_{50}: 27 nM). Dimethyl oxazoles **10c** and **11a** exhibited slightly improved affinity (IC_{50}: 12–16 nM) (see Scheme 3).

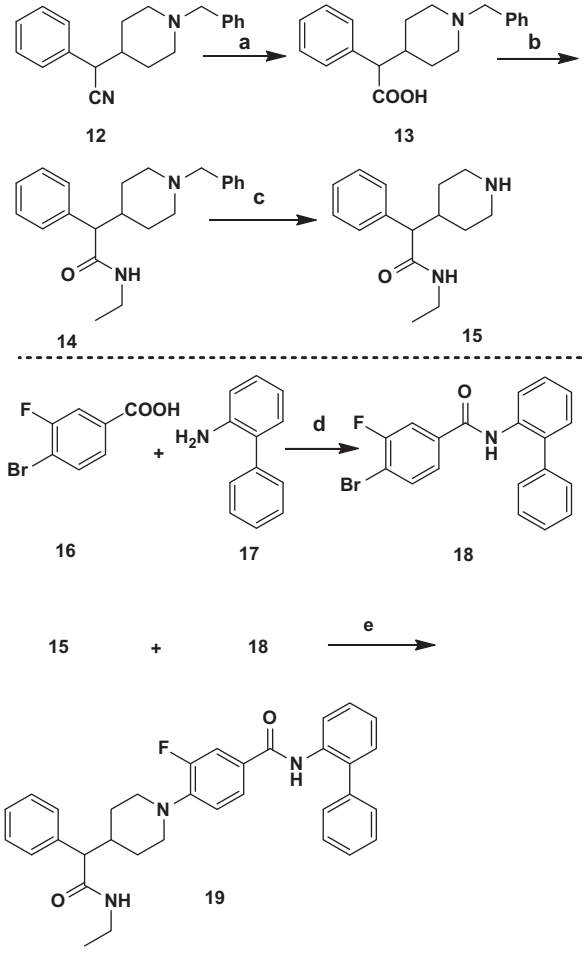
In addition to amides and ureas, the reversed amide was investigated. The reverse amide was prepared from two fragments (**15** and **18**). Compound **12**, benzyl protected 2-phenyl-2-(piperidin-4-yl)acetonitrile was hydrolyzed with 48% HBr providing **13** in good yield (100%). The carboxylic acid **13** reacted with ethylamine, HATU in DMF yielding amide **14** (76%). De-protection of **14** through hydrogenation provided the piperidine fragment **15** (100%). The other fragment bromide **18** was obtained from the amide formation with the acid **16** and the amine **17** (90%). The two fragments (**15** and **18**) were coupled through Pd catalyzed amination yielding the reversed amide **19** (IC_{50}: 45 nM)

Sulfonamide analog **20** was prepared from **6** and (1,1′-biphenyl)-2-sulfonyl chloride (Scheme 4). However, **20** shows no affinity at NPY Y_2 receptor.

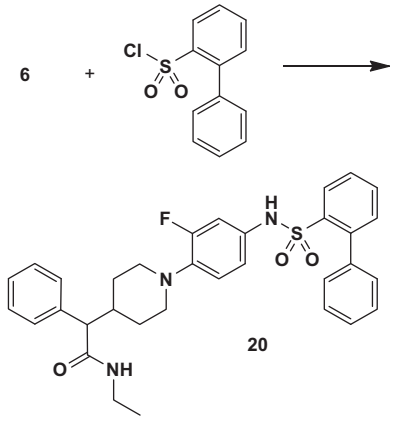
Since compounds **8a**, **8b**, **8c**, **9a**, **9b**, **10a**, **10c**, **11a**, and **19** show good NPY Y_2 binding affinity, their selectivity over MTP was tested using an enzymatic assay.[16,17] The results were summarized in Table 3. Amide **9b** and ureas **10a**, **10c**, **11a** show good selectivity over MTP. Phenyl-pyridine and dimethyl oxazole moieties help improving the selectivity. Urea linkage also favors Y_2 over MTP. The selective compounds were evaluated in NPY Y_2 functional assay[13] and behaved as antagonists (**9b**: $pA_2 = 7.5$; **10c**: $pA_2 = 8.4$; **11a**: $pA_2 = 8.7$).



Scheme 2. Reagents and conditions: R^3NCO, NEt_3, CH_2Cl_2 (60–85%).



Scheme 3. Reagents and conditions: (a) 48% HBr, 105 °C, 100%; (b) H_2NEt, HATU, DMF, 16 °C, 76%; (c) 10%Pd/C, EtOH, 16 °C, 100%; (d) NEt3, HATU, DMF, 16 °C, 90%; (e) $Pd_2(dba)_3$, X-Phos, NaOtBu, $PhCH_3$, 100 °C, 16 h, 18%.



Scheme 4. Reagents and conditions NEt_3, CH_2Cl_2; 70%.

Table 3
Selectivity over MTP

No.	MTP IC_{50} (nM)[16,17]	NPY Y_2 IC_{50} (nM)[18]
8a	13 ± 4	12 ± 4
8b	210 ± 10	24 ± 11
8c	25 ± 5	70 ± 50
9a	7 ± 1	<300
9b	>10,000	37 ± 23
10a	7400	27 ± 7
10c	>10,000	15 ± 2
11a	>10,000	12 ± 6
19	33 ± 2	45 ± 15

Table 4
Microsomal stability (remaining after 15 min in human and rat)

No.	Microsomal stability (Hu)	Microsomal stability (Rat)
1	1.4% Rmn	1.7% Rmn
9a	73% Rmn	98% Rmn
9b	5.3% Rmn	3.3% Rmn
11a	1.5% Rmn	17% Rmn

Amides and ureas with piperidinyl core show improved microsomal stability (Table 4). Amide **9a** with piperidinyl core exhibits much better microsomal stability (human: 73% remaining after 15 min; rat: 98% remaining after 15 min) comparing to **1** (human: 1.4% remaining after 15 min; rat 1.7% remaining after 15 min), however it is less selective over MTP. Amide **9b** and urea **11a** with piperidinyl core also show improved microsomal stability (human: 5.3–1.5% remaining after 15 min; rat: 3.3–17% remaining after 15 min) comparing to **1**. However they are less stable than compound **9a**. Both **9b** and **11a** has great selectivity over MTP.

Due to still less favorable microsomal stability, further in vivo evaluations of **9b** and **11a** were limited and performed through subcutaneous administration. After subcutaneous administration in rat, **9b** crossed the blood brain barrier and occupied NPY Y_2 receptors (approximately 50% at 10 mg/Kg, experiments were performed according to the procedure described in Ref. 13). However, ureas **10c** and **11a** showed no brain penetration and consequently no NPY Y_2 receptor occupancy.

In summary, a series of compounds with a piperidinyl core were evaluated for NPY Y_2 antagonism. Different amides and amide analogs were investigated. We identified new heterocyles: dimethyl oxazoles showing excellent selectivity over MTP (**9b**, **10c**, **11a**). Ureas show good selectivity over MTP comparing amides and reverse

amides. Amides and ureas with piperidinyl core showed improved microsomal stability. These new finding certainly will help us to design better NPY Y_2 antagonists in the future. Potent and selective ureas **10c** and **11a** showed no brain penetration, and thus they are potentially useful tool compounds to help understand the roles of the NPY Y_2 receptors in the periphery.

References and notes

1. Tatemoto, K. *Proc. Natl. Acad. Sci. U.S.A.* **1982**, *79*, 5485.
2. Blomqvist, A. G.; Herzog, H. *Trends Neurosci.* **1997**, *20*, 294.
3. Kaga, T.; Fujimiya, M.; Inui, A. *Peptides* **2001**, *22*, 501.
4. Redrobe, J. P.; Dumont, Y.; Herzog, H.; Quirion, R. *Behav. Brain Res.* **2003**, *141*, 251.
5. Sato, N.; Ogino, Y.; Mashiko, S.; Ando, M. *Expert Opin. Ther. Pat.* **2009**, *19*, 1401.
6. Baldock, P. A.; Sainsbury, A.; Couzens, M.; Enriquez, R. F.; Thomas, G. P.; Gardiner, E. M.; Herzog, H. *J. Clin. Invest.* **2002**, *109*, 915.
7. Rimondini, R.; Thorsell, A.; Heilig, M. *Neurosci. Lett.* **2005**, *375*, 129.
8. Doods, H.; Gaida, W.; Wieland, H. A.; Dollinger, H.; Schnorrenberg, G.; Esser, F.; Engel, W.; Eberlein, W.; Rudolf, K. *Eur. J. Pharmacol.* **1999**, *384*, R3.
9. Andres, C. J.; Zimanyi, I. A.; Deshpande, M. S.; Iben, L. G.; Grant-Young, K.; Mattson, G. K.; Zhai, W. *Bioorg. Med. Chem. Lett.* **2003**, *13*, 2883.
10. Lunniss, G. E.; Barnes, A. A.; Barton, N.; Biagetti, M.; Bianchi, F.; Blowers, S. B.; Caberlotto, L.; Emmons, A.; Holmes, I. P.; Montanari, D.; Norris, R.; Walters, D. J.; Watson, S. P. *Bioorg. Med. Chem. Lett.* **2009**, *19*, 4022.
11. Brothers, S. P.; Saldanha, S. A.; Spicer, T. P.; Cameron, M.; Mercer, B. A.; Chase, P.; McDonald, P.; Wahlestedt, C.; Hodder, P. S. *Mol. Pharmacol.* **2010**, *77*, 46.
12. Jablonowski, J. A.; Chai, W.; Li, X.; Rudolph, D. A.; Murray, W. V.; Youngman, M. A.; Dax, S. L.; Nepomuceno, D.; Bonaventure, P.; Lovenberg, T. W.; Carruthers, N. I. *Bioorg. Med. Chem. Lett.* **2004**, *14*, 1239.
13. Bonaventure, P.; Nepomuceno, D.; Mazur, C.; Lord, B.; Rudolph, D. A.; Jablonowski, J. A.; Carruthers, N. I.; Lovenberg, T. W. *J. Pharmacol. Exp. Ther.* **2004**, *308*, 1130.
14. Swanson, D. M.; Wong, V. D.; Jablonowski, J. A.; Shah, C.; Rudolph, D. A.; Dvorak, C. A.; Seierstad, M.; Dorark, L. K.; Morton, K.; Nepomuceno, D.; Atack, J. R.; Bonaventure, P.; Lovenberg, T. W.; Carruthers, N. I. *Bioorg. Med. Chem. Lett.* **2011**, *21*, 5552.
15. Seierstad, M.; Bonaventure, P.; Dvorak, L.; Lord, B.; Miller, K. L.; Motley, S. T.; Nepomuceno, D.; Chai, W.; Dvorak, C. A.; Jablonowski, J.; Rudolph, D. A.; Shah, C. R.; Swanson, D. M.; Wong, V. D.; Axe, F. U.; Lovenberg, T. W.; Carruthers, N. I. *Abstracts of Papers*, 234th ACS National Meeting, Boston, MA, Aug 19–23, 2007; USA.
16. Meerpoel, L.; Roevens, P.; Walter, M.; Backx, L. J. J.; Van der Veken, L. J. E.; Viellevoye, M. PCT Int. Appl. WO 2002020501, A2 20020314, 2002.
17. 18. Meerpoel, L.; Viellevoye, M. PCT Int. Appl. WO 2002081460, A1 20021017, 2002.
18. NPY Y2 binding affinity was determined as detailed in Bonaventure et al *J. Pharmacol. Exp. Ther.* **2004**, *308*, 1130. IC_{50} values are the mean of 3–10 determinations followed by SEM. All compounds were characterized by ^1H NMR and MS.