Bioorganic & Medicinal Chemistry Letters 23 (2013) 1519–1521

Contents lists available at SciVerse ScienceDirect

Bioorganic & Medicinal Chemistry Letters

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

Synthesis and biological evaluation of 5-nitropyrimidine analogs with azabicyclic substituents as GPR119 agonists

Zunhua Yang [a,b], Yuanying Fang [a], Tuan-Anh N. Pham [a], Jongkook Lee [a], Haeil Park [a,*]

[a] College of Pharmacy of Kangwon National University, Chuncheon 200-701, Republic of Korea
[b] College of Pharmacy, Jiangxi University of Traditional Chinese Medicine, Nanchang 330004, China

ARTICLE INFO

Article history:
Received 29 October 2012
Revised 4 December 2012
Accepted 8 December 2012
Available online 20 December 2012

Keywords:
GPR119 agonist
Type 2 diabetes
5-Nitropyrimidines
Azabicyclic alcohols
Azabicyclic amines

ABSTRACT

5-Nitropyrimidine analogs substituted with conformationally restricted azabicyclic amines and alcohols were prepared and evaluated their agonistic activity against human GPR119. The analogs bearing *endo*-azabicyclic amines and alcohols (**7**, **8**, **11**, and **12**) exhibited full agonistic activities while the analogs with *exo*-azabicyclic amines and alcohols were proved as partial agonists (**9**, **10**, **13**, and **14**) regardless of their EC_{50} values. 5-Nitropyrimidine analogs with (2-fluoro-4-methylsulfonyl)phenylamino group (**8**, **10**, **12**, **14**) showed more potent GPR119 activation activities than the analogs without fluorine in all cases (**7**, **9**, **11**, **13**).

Type 2 diabetes is a complex metabolic disorder and is characterized by a high blood glucose level that caused by insulin resistance or insufficient insulin secretion, accounting for approximately 90% of all cases of diabetes.[1] Although the number of patients with type 2 diabetes that successfully achieve target glucose levels is steadily improving, a substantial number of subjects continue to fall short of acceptable treatment goals, leaving them at high risk of development of diabetes-associated complications such as hypoglycemia, weight gain, bone fractures, etc.[1–3] Therefore, new drugs with novel mode of action that exhibit improved efficacy and safety relative to current available medications are clearly needed.

GPR119 is a G-protein coupled receptor predominantly expressed on pancreatic beta cells and intestinal enteroendocrine cells. GPR119 agonists such as oleoylethanolamine (OEA), lysophosphatidylcholine, *N*-oleoyldopamin and olvanil were reported to stimulate glucose-dependent insulin secretion in vitro and lower elevated blood glucose level in vivo. Furthermore, it was demonstrated that they stimulated the release of the incretin (GLP-1 and GIP).[4–6] Therefore, GPR119 agonist has emerged as a promising target for the treatment of type 2 diabetes and obesity by improving glucose homoeostasis while concurrently slowing gastric emptying, reducing food intake and promoting weight loss.[4–6]

Recently, efforts have been made toward development of antidiabetic agents for type 2 diabetes targeting GPR119. Diverse fused aromatic heterocycles were found in the structures of GPR119 agonists under development (Fig. 1).[6–11] Also piperidine scaffold was attached to the diverse aromatic heterocycles in most cases. These results implied that piperidine scaffold plays an important role for bioactivity. Also 2-fluoro-4-methanesulfonylamine fragment is very common in the structures of developing GPR119 agonists as shown in the structures of APD-668 and AR-231453.

In our efforts to discover pharmacologically superior GPR119 agonists, pyrimidine derivative AR-231453 was chosen as a lead structure and planned to replace the piperidine ring to the conformationally restricted azabicyclic ring to produce new pyrimidine analogs (Fig. 2). One of our strategies included the incorporation of ethylene to conformationally constrain the piperidine ring, one of the more common motifs found in GPR119 agonist structures. It was suggested from a precedent literature that the piperidine carbamate region seemed to make pharmacologically very important H-bond with the receptor.[12] Therefore, it is surmised that sterical changes in this region cause significant pharmacological changes. Herein we report synthesis of 5-nitropyrimidine analogs with conformation restricted azabicyclic amines/alcohols as GPR119 agonists for the treatment of type 2 diabetes.

5-Nitropyrimidine analogs were prepared from key intermediates **1** and **2** following the procedures and conditions as shown in Scheme 1. Reaction of 4,6-dichloropyrimidine[13] with 4-(methylthio)aniline or 2-fluoro-4-(methylthio)aniline[14] in DMF followed by oxidation with *m*CPBA afforded compounds **1** and **2** in 58% and 56%, respectively. Coupling reactions of the intermediates (**1**, **2**) and *endo*-azabicyclic amine **3** with diisopropylethylamine

* Corresponding author. Tel.: +82 33 250 6920; fax: +82 33 255 7865.
E-mail address: haeilp@kangwon.ac.kr (H. Park).

0960-894X/$ - see front matter © 2012 Elsevier Ltd. All rights reserved.
http://dx.doi.org/10.1016/j.bmcl.2012.12.011

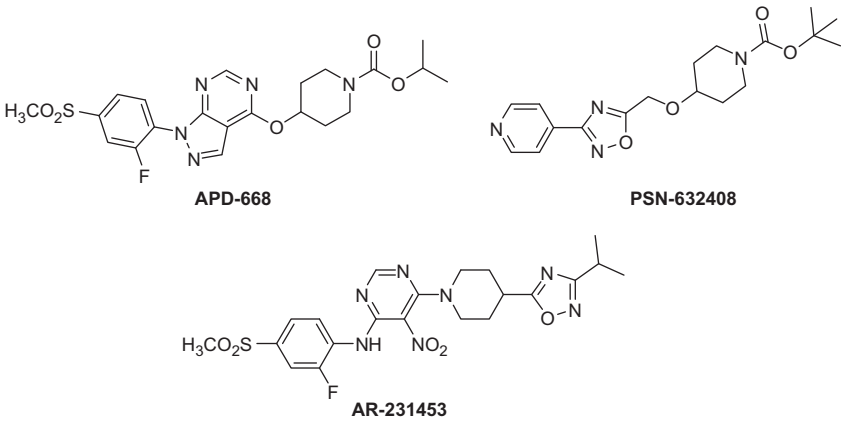


Figure 1. Structures of selected GPR119 agonists under clinical development.

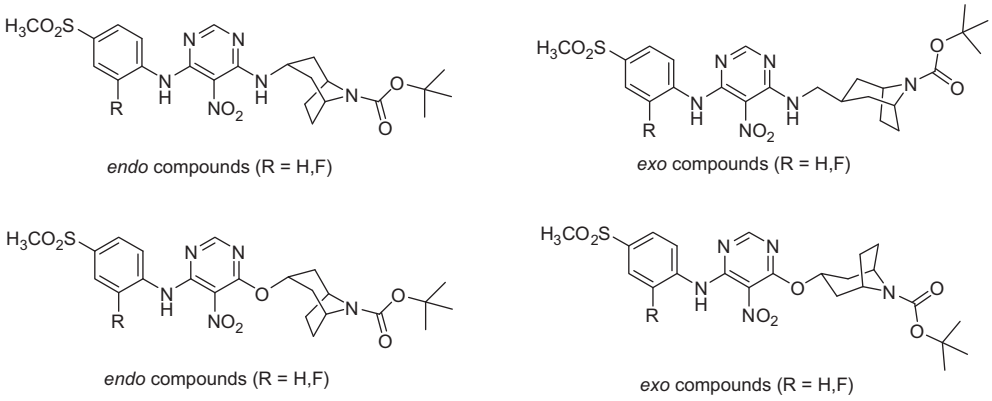
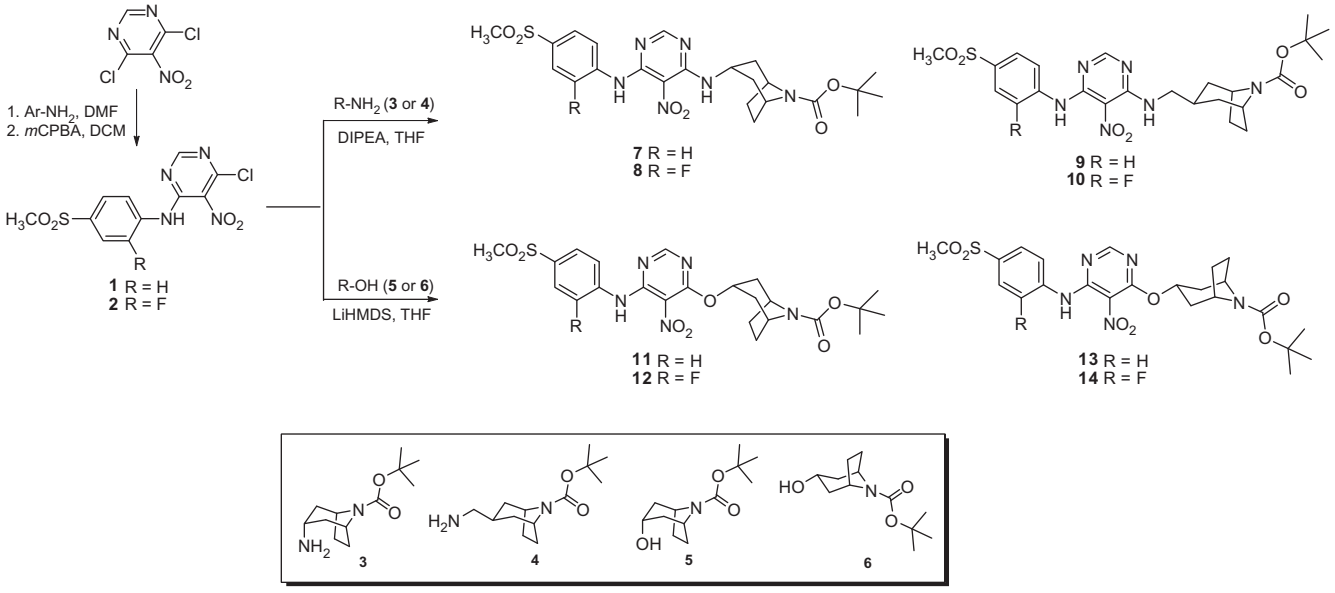


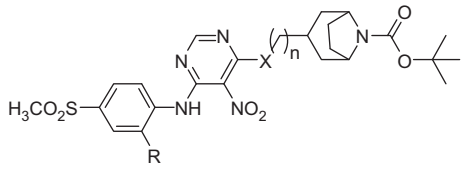
Figure 2. Structures of target GPR119 agonists.



Scheme 1. Synthetic pathways for target GPR119 agonists.

(DIPEA) in THF yielded products (**7**, **8**) in 81% and 89%, respectively. Reactions of the intermediates (**1**, **2**) and *exo*-azabicyclic amine **4** with diisopropylethylamine (DIPEA) in THF yielded products (**8**, **9**) in 83% and 80%, respectively. Coupling reactions of the intermediates (**1**, **2**) and *endo*-azabicyclic alcohol **5** with lithium hexamethyldisilazide (LiHMDS) in THF yielded products (**11**, **12**) in 68% and 65%, respectively. Also coupling reactions of the intermediates (**1**, **2**) and *exo*-azabicyclic alcohol **6** yielded products (**13**, **14**) in 71% and 75%, respectively. Azabicyclic amines (**3** and **4**) and alcohols (**5** and **6**) were prepared following known methods in literatures.[15]

Table 1
In vitro GPR119 agonistic activities of 5-nitropyrimidine analogs (**7**–**14**)



(R = H,F; X = O,N ; n = 0,1)

Compound	R	X	*n*	hGPR119 activity	
				EC_{50}[a] (nM)	% Max[b]
7	H	N	0	11.5	139
8	F	N	0	1.5	99
9	H	N	1	>10,000	41
10	F	N	1	6.7	53
11	H	O	0	4.9	99
12	F	O	0	1.5	116
13	H	O	0	14.5	69
14	F	O	0	1.4	65
OEA				2200	100

[a] Concentration for 50% cAMP stimulation of OEA.
[b] cAMP stimulation % compared to maximal effect of OEA.

Compounds **7**–**14** were evaluated their abilities to activate the human GPR119 receptor in a cell-based cAMP assay and, which were expressed in EC_{50} and % max values. The EC_{50} values represent the concentration of the tested compounds for 50% cAMP of oleylethanolamine (OEA), while the % max values present the relative response (%) of the tested compounds compared to the maximal effect of OEA.[16]

As shown in Table 1, most 5-nitropyrimidine analogs possessing azabicyclic amines/alcohols exhibited more potent GPR119 agonistic activity than OEA, however, some analogs were identified as partial agonists (**9**, **10**, **13**, **14**). All the 5-nitropyrimidine analogs with (2-fluoro-4-methylsulfonyl)phenylamino group (**10**, **12**, **14**) showed more potent GPR119 activation activities than the analogs with 4-(methylsulfonyl)phenylamino group (**9**, **11**, **13**). One of very interesting phenomena was observed that the analogs (**7**, **8**, **11**, **12**) with *endo*-azabicyclic amine (**3**) and alcohol (**5**) were proved as full agonists while the analogs (**9**, **10**, **13**, **14**) with *exo*-azabicyclic amine (**4**) and alcohol (**6**) were proved as partial agonists regardless of their EC_{50} values. It may be caused that the analogs with *endo*-azabicyclic amine or alcohol are constrained to the agonist conformation and are full agonists.[12]

In summary, in the present article, we have reported the synthesis of eight 5-nitropyrimidine analogs possessing conformationally restricted azabicyclic amines and alcohols and biological evaluation of their abilities to activate the human GPR119 receptor in a cell-based cAMP assay, which are expressed in EC_{50} and % max values. The analog **7** exhibited maximum agonistic activity (139% max) with slightly weak EC_{50} value (11.5 nM) while the analog **12** exhibited quite good agonistic activity (116% max) with strong EC_{50} value (1.5 nM). Our present results also revealed that analogs with fluorine substituted phenylamino group showed more potent receptor agonistic activity than analogs without fluorine in all cases and analogs with *endo*-azabicycle moiety showed full agonistic activities while analogs with *exo*-azabicycle moiety showed partial agonistic activities. Our present results are completely in accordance with those from a precedent paper.[12] These preliminary results encourage us to search diverse conformation restricted azabicyclic motifs and also heteocycle rings as congeners of pyrimidine ring. The follow-up studies and results will be reported in due course.

Acknowledgements

The authors thank to Dr. Moon Ho Son (Dong-A Pharm. Co. Ltd.) for hGPR119 cAMP reporter assay. The authors also thank to Pharmacal Research Institute and Central laboratory of Kangwon National University for the use of bioassay facilities and analytical instruments.

References and notes

1. Schmeltz, L.; Metzger, B. In Williams, M., Ed., 2nd ed.; Comprehensive Medicinal Chemistry; Elsevier: Oxford, 2007; Vol. 6, pp 417–458.
2. Drucker, D. J.; Sherman, S. I.; Gorelick, F. S.; Bergenstal, R. M.; Sherwin, R. S.; Buse, J. B. *Diabetes Care* **2010**, *33*, 428.
3. Ahren, B. *Nat. Rev. Drug Disc.* **2009**, *8*, 369.
4. Overton, H. A.; Babbs, A. J.; Doel, S. M.; Fyfe, M. C. T.; Gardner, L. S.; Griffin, G.; Jackson, H. C.; Procter, M. J.; Rasamison, C. M.; Tang-Christensen, M.; Widdowson, P. S.; Williams, G. M.; Reynet, C. *Cell Metab.* **2006**, *3*, 167.
5. Overton, H. A.; Fyfe, M. C. T.; Reynet, C. *Br. J. Pharmacol.* **2008**, *153*, S76.
6. Jones, R. M.; Leonard, J. N. *Annu. Rep. Med. Chem.* **2009**, *44*, 149.
7. Hansen, H. S.; Rosenkilde, M. M.; Holst, J. J.; Schwartz, T. W. *Trends Pharmacol. Sci.* **2012**, *33*, 374.
8. Semple, G.; Fioravanti, B.; Pereira, G.; Calderon, I.; Uy, J.; Choi, K.; Xiong, Y.; Ren, A.; Morgan, M.; Dave, V.; Thomsen, W.; Unett, D. J.; Xing, C.; Bossie, S.; Carroll, C.; Chu, Z.; Grottick, A. J.; Hauser, E. K.; Leonard, J.; Jones, R. M. *J. Med. Chem.* **2008**, *51*, 5172.
9. Wu, Y.; Kuntz, J. D.; Carpenter, A. J.; Fang, J.; Sauls, H. R.; Gomez, D. J.; Ammala, C.; Xu, Y.; Hart, S.; Tadepalli, S. *Bioorg. Med. Chem. Lett.* **2010**, *20*, 2577.
10. Xia, Y.; Chackalamannil, S.; Greenlee, W. J.; Jayne, C.; Neustadt, B.; Stamford, A.; Vaccaro, H.; Xu, X.; Baker, H.; O'Neill, K.; Woods, M.; Hawes, B.; Kowalski, T. *Bioorg. Med. Chem. Lett.* **2011**, *21*, 3290.
11. Semple, G.; Ren, A.; Fioravanti, B.; Pereira, G.; Calderon, I.; Choi, K.; Xiong, Y.; Shin, Y. J.; Gharbaoui, T.; Sage, C. R.; Morgan, M.; Xing, C.; Chu, Z. L.; Leonard, J. N.; Grottick, A. J.; Al-Shamma, H.; Liang, Y.; Demarest, K. T.; Jones, R. M. *Bioorg. Med. Chem. Lett.* **2011**, *21*, 3134.
12. McClure, K. F.; Darout, E.; Guimarães, C. R.; DeNinno, M. P.; Mascitti, V.; Munchhof, M. J.; Robinson, R. P.; Kohrt, J.; Harris, A. R.; Moore, D. E.; Li, B.; Samp, L.; Lefker, B. A.; Futatsugi, K.; Kung, D.; Bonin, P. D.; Cornelius, P.; Wang, R.; Salter, E.; Hornby, S.; Kalgutkar, A. S.; Chen, Y. *J. Med. Chem.* **1948**, *2011*, 54.
13. Latli, B.; Jones, P. J.; Krishnamurthy, D.; Senanayake, C. H. *J. Labelled Compd. Radiopharm.* **2008**, *51*, 54.
14. Zhu, W.; Ma, D. *J. Org. Chem.* **2005**, *70*, 2696.
15. For preparation of azabicyclic amines **3** and **4** see: (a) Berdini, V.; Cesta, M. C.; Curti, R.; D'Anniballe, G.; Bello, N. D.; Nano, G.; Nicolini, L.; Topai, A.; Allegretti, M. *Tetrahedron* **2002**, *58*, 5669; (b) Kazmierski, W. M.; Aquino, C.; Chauder, B. A.; Deanda, F.; Ferris, R.; Jones-Hertzog, D. K.; Kenakin, T.; Koble, C. S.; Watson, C.; Wheelan, P.; Yang, H.; Youngman, M. *J. Med. Chem.* **2008**, *51*, 6538; For preparation of azabicyclic alcohols **5** and **6** see: (c) Nagase, T.; Takahashi, T.; Sasaki, T.; Nagumo, A.; Shimamura, K.; Miyamoto, Y.; Kitazawa, H.; Kanesaka, M.; Yoshimoto, R.; Aragane, K.; Tokita, S.; Sato, N. *J. Med. Chem.* **2009**, *52*, 4111.
16. *Human GPR119 cAMP reporter assay:* HEK293 cells (4×10^3 cells/well) were seeded on 96 half-well plates and incubated for 24 h. The cells were transected with GPR119 expression plasmid (OriGene Technologies, Inc., USA) using Lipofectamine and Plus reagent (Life Technologies Corporation., USA). After 24 h, transfected cells were incubated with compounds dissolved in assay buffer (KRBH buffer containing 0.1% BSA and 500 μM 3-isobutyl-1-methylxanthine) for 60 min at 37 °C. Subsequently, cells were harvested with lysis buffer (50 mM phosphate buffer containing 1 M KF and 1.25% Triton X-100, pH 7.0) for 10 min at room temperature and the assay was performed using the cAMP homogeneous time-resolved fluorescence kit (CIS bio international, France).