Bioorganic & Medicinal Chemistry Letters 18 (2008) 5032–5036

Contents lists available at ScienceDirect

Bioorganic & Medicinal Chemistry Letters

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

Benzimidazole-substituted (3-phenoxypropyl)amines as histamine H3 receptor ligands

Robert Aslanian *, Xiaohong Zhu, Henry A. Vaccaro, Neng-Yang Shih, John J. Piwinski, Shirley M. Williams, Robert E. West, Jr.

The Schering Plough Research Institute, 2015 Galloping Hill Road, Kenilworth, NJ 07033-1300, USA

ARTICLE INFO

Article history:
Received 5 June 2008
Revised 31 July 2008
Accepted 4 August 2008
Available online 7 August 2008

Keywords:
Histamine
H3

ABSTRACT

A series of non-imidazole histamine H_3 receptor antagonists based on the (3-phenoxypropyl)amine motif, which is a common pharmacophore for H_3 antagonists, has been identified. A preliminary SAR study around the amine moiety has identified **8a** as a potent H_3 antagonist possessing a good pharmacokinetic profile in the rat.

The endogenous amine, histamine, is a ligand for four distinct histaminergic receptor subtypes, namely the H_1, H_2, H_3, and H_4 receptors.[1] H_1 receptors are located in the CNS, and on skin and smooth muscle in the airways; H_2 receptors are located in the gastrointestinal tract. Antagonists of the H_1 and H_2 receptors are well-known therapeutic agents, and have been used clinically for many years for the treatment of allergic diseases and ulcers, respectively. H_3 receptors are located primarily in the CNS where they act as both autoreceptors for histamine as well as heteroreceptors for other neurotransmitters. Evidence from in vivo studies in animals indicates that H_3 receptor ligands may be useful for the treatment of a variety of CNS-related disorders like ADHD, Alzheimer disease, sleep disorders, and neuropathic pain.[2] Additionally, given the known role of central histamine in the control of appetite, H_3 receptor ligands are also reported to be active in animal models of obesity.[2,3] The H_4 receptor is located on various hematopoietic cells and functions in chemotaxis of these cells.[4] There are currently no marketed drugs that act at either the H_3 or H_4 receptors, although several companies have entered clinical trials with H_3 antagonists/inverse agonists for the treatment of cognition, pain, and narcolepsy.[5] Recently, positive clinical data in a small clinical trial for narcolepsy have been reported.[6]

Structurally, the first generation of H_3 ligands were analogs of histamine in that they were based on a 4-substituted imidazole motif. Typical examples of agonists and antagonists are given in Figure 1. These compounds were useful tools in delineating some of the pharmacology of the H_3 receptor, but in general imidazole derivatives make poor drug candidates due to their propensity to inhibit numerous mammalian CYP450 isozymes,[7] and in the case of a CNS drug, their poor brain penetration. Therefore, a significant synthetic effort was undertaken in academia and industry to identify non-imidazole-derived H_3 antagonists. Although the natural product Aplysamine-1, identified by scientists at Harbor Branch Oceanographic Institute and Schering-Plough, was known as a weak H_3 receptor ligand,[8] the first real breakthrough came from Ganellin and co-workers who identified the phenoxyalkyl amine motif of Aplysamine-1 as a potent H_3 pharmacophore (UCL 1972, Fig. 2).[9] Subsequently, a large number of groups have utilized this pharmacophore as the basis of their own programs (Fig. 2).[2a] More recently, other H_3 pharmacophores have been described, but none have been as thoroughly examined as the phenoxyalkyl amine.[10,11]

When examining the structures of compounds based on this pharmacophore described in the primary and patent literature, we were struck not only by the similarity of many of the structures but also by the apparent promiscuity of the receptor toward moieties on the phenyl ring. Simple moieties like the nitro group as well as basic groups, fused rings, and heterocycles are all tolerated. Based on a series of benzimidazole-substituted analogs previously prepared in our laboratories, we decided to investigate a series of compounds in which the phenyl ring was substituted by an N-linked 5-fluorobenzimidazole moiety (Fig. 3).[12] The results are described herein.

The syntheses of the analogs in Tables 1 and 2 are described in Schemes 1 and 2.[13] Initially, we decided to focus on analogs incorporating a 2-pyridyl, thioether, or ether moiety at the 2-position of the benzimidazole ring based on our previous experience.[12] Briefly, the benzimidazole moiety is constructed by reacting amine **1** with

* Corresponding author. Tel.: +1 908 740 3514; fax: +1 908 740 7152.
E-mail address: robert.aslanian@spcorp.com (R. Aslanian).

0960-894X/$ - see front matter © 2008 Elsevier Ltd. All rights reserved.
doi:10.1016/j.bmcl.2008.08.008

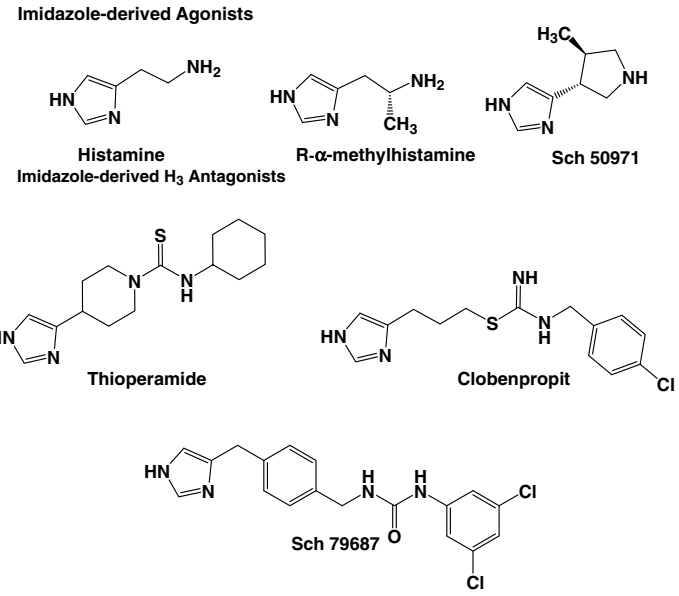


Figure 1. Examples of imidazole-derived agonists and antagonists.

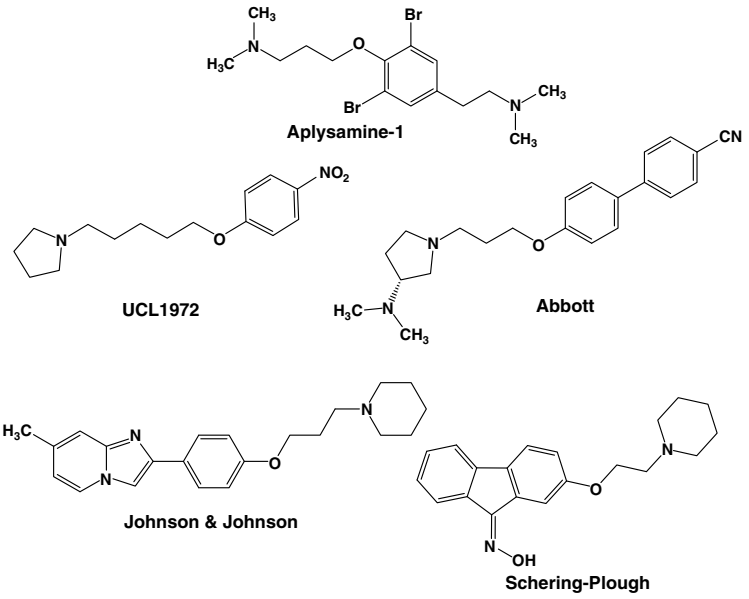


Figure 2. Examples of non-imidazole-derived H_3 antagonists.

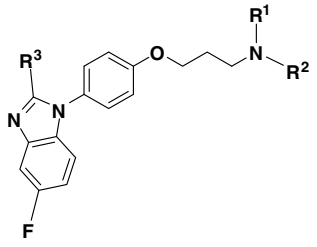


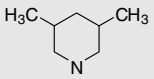
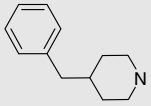
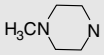
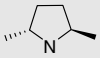
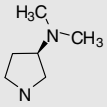
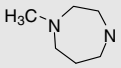
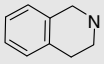
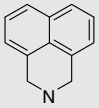
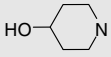
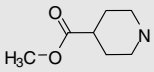
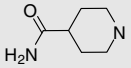
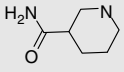
Figure 3.

difluoronitrobenzene **2** to yield **3**. Reduction of the nitro group of **3** gave compound **4,** which was coupled with picolinic acid to give amide **5**. Heating **5** in acetic acid results in ring closure to give benzimidazole **6**, which was alkylated on the phenol to give key intermediate **7**. Chloride **7** was easily converted to a variety of amines **8** utilizing parallel synthesis techniques.

The synthesis of analogs incorporating either an ether or a thioether in the 2-position of the benzimidazole moiety is given in Scheme 2.

Treatment of phenol **9** with base and 1-bromo-3-chloropropane followed by displacement of the chlorine with piperidine gave **11** in excellent yield for the two steps. Reduction of the nitro group of **11** with Ra-Ni and reaction with **2** gave compound **12**. Reduction of the nitro group, again with Ra-Ni, gave the amine which was reacted with either thiocarbonyl diimidazole (X=S) or carbonyl diimidazole (X=O) to give **14a** and **14b**. Treatment of **14a** or **14b** with base and an alkyl halide gave the ethers **15a** and **15b**. Human H_3 binding data for analogs **8**, **14**, and **15** are given in Tables 1 and 2.[14]

Table 1
Human H_3 binding data for benzimidazoles **8**[14]

Benzimidazole	Amine	H_3 K_i[a] (nM)
8a		1.2
8b		11
8c		3.4
8d		2.0
8e		13
8f		1.2
8g		5.1
8h		4.8
8i		20
8j		5.3
8k		3.6
8l		107
8m		713
8n		1.8
8o		16
8p		20
8q		11
8r		101

[a] H3 binding K_i values are the average of at least two independent determinations.

Table 2
H_3 binding data for **14** and **15**[14]

Compound	X or RX	H_3 K_i[a] (nM)
14a	O	8.7
14b	S	8.0
15a	SCH_2CH_3	2.6
15b	OCH_2CH_3	15

[a] H3 binding K_i values are the average of at least two independent determinations.

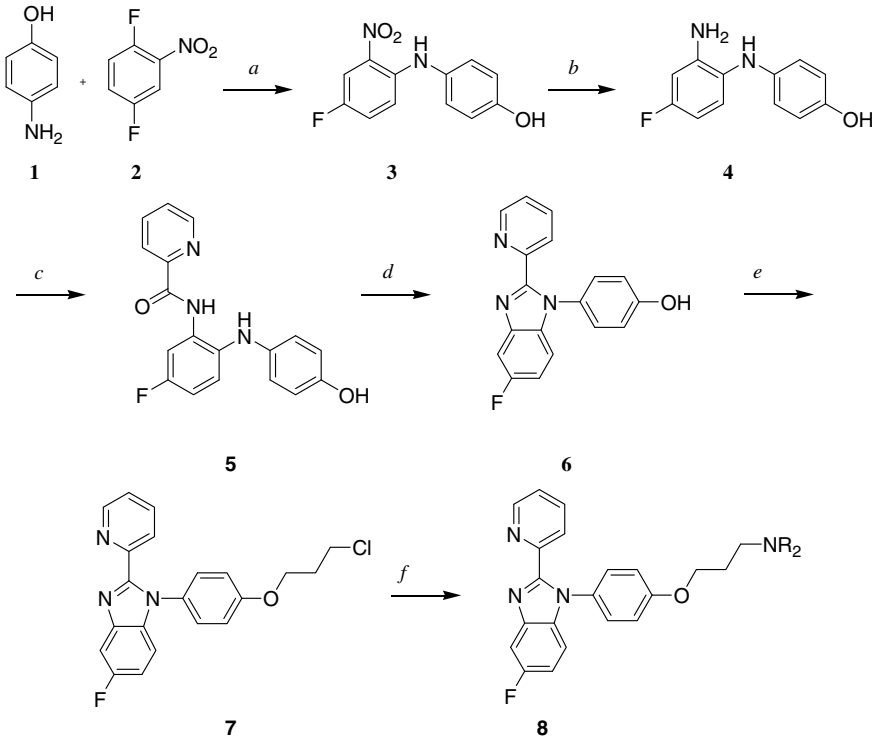
A number of conclusions regarding the SAR of this series of benzimidazole-derived H_3 ligands can be drawn from these data. First, five-, six-, and seven-membered monocyclic amines give good to excellent H_3 binding activity, and a second heteroatom is tolerated within the ring (entries **8a**–**k**). Bulkier, fused bi- and tricyclic amines, however, are less active (entries **8l** and **8m**) although some steric bulk is tolerated (**8d**). It is interesting to note that a simple methylamine substituent does not result in good binding affinity (**8r**). Polar substituents like amino, hydroxyl, ester, and amide are tolerated on the pyrrolidine and piperidine ring (entries **8h** and **8n**–**8q**). The position of the substituent on the ring does not appear to be critical to the binding affinity in the case that was examined (**8p** vs **8q**).

The nature of the substituents on the benzimidazole moiety does not seem to play a major role in the binding profile. Heteroatomic, heteroaroyl, and ether substituents are tolerated at the 2-position of the benzimidazole ring (Table 2). For example, analogs **14a** and **14b**, featuring either oxygen or sulfur in the 2-position, or analogs **15a** and **15b** are good ligands for the H_3 receptor. Taken in aggregate, these data are consistent with the idea that the phenoxyalkyl amine H_3 pharmacophore used in this study is, in general, very tolerant of different substituents on the phenyl ring.

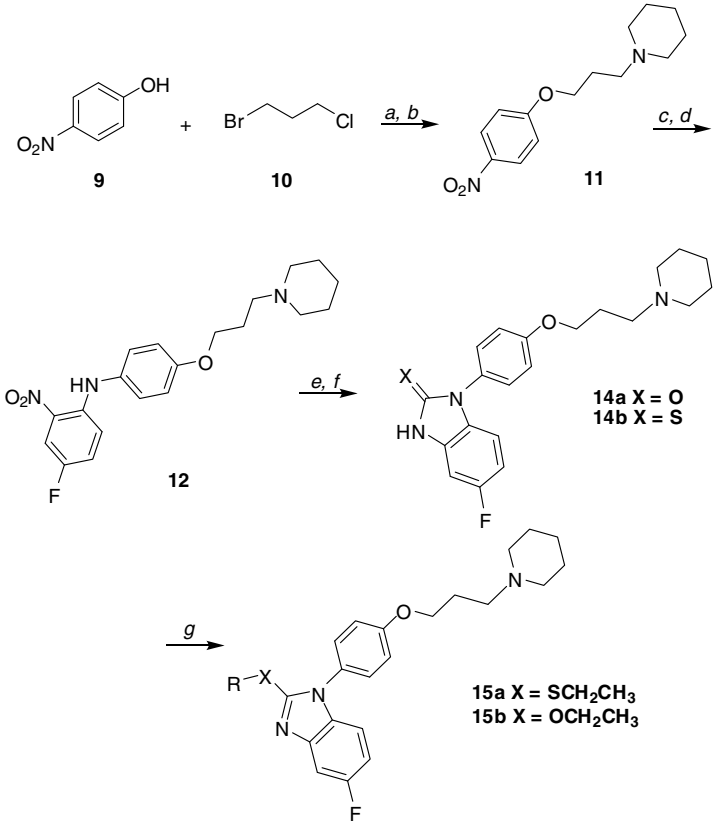
In addition to H_3 binding activity, hERG activity was measured using a high-throughput rubidium efflux assay.[15] Representative data are given in Table 3. In general, hERG activity as measured by this assay is quite high regardless of the nature of the amine present on the terminal position. This observation is consistent with other similar series of H_3 ligands incorporating this pharmacophore that also displays hERG activity.[2b] Interestingly, introduction of polar substituents, an approach that has frequently demonstrated a positive impact on hERG activity, could improve the hERG profile of some analogs in this series as well (Table 3, compounds **8h** and **8n**).

Because of its excellent activity at the human H_3 receptor, **8a** was further profiled. Compound **8a** had a K_i of 1.8 nM when tested against the mouse H_3 receptor, and its in vitro functional activity as measured in a human cAMP assay was 0.1 nM.[16] It was a full antagonist. It did not inhibit CYP3A4 or 2D6 when tested at concentrations up to 30 μM.[17] Furthermore, **8a** possessed a reasonable oral pharmacokinetic profile in a high-throughput rat pharmacokinetic assay: AUC = 2027 h ng/mL; C_{max} = 393 ng/mL (10 mg/kg in methyl cellulose, n = 2, 0–6 h time points).[18]

In conclusion, a new series of H_3 ligand based on a well-known H_3 pharmacophore, the phenoxyalkyl amine, have been identified. Many of these analogs possess excellent H_3 binding affinity, but like many analogs that incorporate this motif, they also have the potential to interact with the hERG channel. This series also helps to demonstrate the promiscuous nature of the receptor toward this pharmacophore.



Scheme 1. Reagents and conditions: (a) Dioxane, reflux, 64%; (b) Ra-Ni, H_2, MeOH, 83%; (c) picolinic acid, DCC, HOBT, CH_2Cl_2, 48%; (d) Acetic acid, 120 °C, 100%; (e) 1-bromo-3-chloropropane, K_2CO_3, acetone, reflux, 93%; (f) R_2NH, CH_3CN, *i*-Pr_2NEt, 80 °C, 12–98%.



Scheme 2. Reagents and conditions: (a) Acetone, K_2CO_3, R.T., 97%; (b) Piperidine, *n*-butanol, NaI, Na_2CO_3, 100 °C, 100%; (c) Ra-Ni, H_2, MeOH, 85%; (d) **2**, dioxane, reflux, 68%; (e) Ra-Ni, H_2, MeOH, 94%; (f) Carbonyl diimidazole or thiocarbonyl diimidazole, 70 °C, 100% for **14a**, 98% for **14b**; (g) CH_3CH_2I, DMF, K_2CO_3, 60% for **15a**, 65% for **15b**.

Table 3
Rubidium efflux data for selected analogs[a]

Compound	Rb efflux (%)
8a	70
8c	62
8h	26
8i	42
8l	94
8n	16
8o	49

[a] Compounds were tested at a concentration of 5 μg/mL.

Acknowledgments

We thank the Drug Metabolism and Pharmacokinetics group of the Schering-Plough Research Institute, for providing the in vitro and in vivo pharmacokinetic data, and Steve Sorota for providing the hERG data.

References and notes

1. For a recent review of the histaminergic receptor system, see: Parsons, M. E.; Ganellen, C. R. *Br. J. Pharmacol.* **2006**, *147*, S127.
2. Recent reviews of the H3 receptor: (a) Celanire, S.; Wijtmans, M.; Telaga, P.; Leurs, R.; de Esch, I. J. P. *Drug Discov. Today* **2005**, *10*, 1613; (b) Hancock, A. A. *Biochem. Pharmacol.* **2006**, *71*, 1103; (c) Esbenshade, T. A.; Fox, G. B.; Cowart, M. D. *Mol. Interventions* **2006**, *6*, 77; (d) Bongers, G.; Bakker, R. A.; Leurs, R. *Biochem. Pharmacol.* **2007**, *73*, 1195.
3. (a) Hancock, A. A.; Brune, M. E. *Expert Opin. Invest. Drugs* **2005**, *14*, 223; (b) Malmlof, K.; Hastrup, S.; Schellerup Wulff, B.; Hansen, B.; Peschke, B.; Jeppesen, C.; Hohlweg, R.; Rimvall, K. *Biochem. Pharmacol.* **2007**, *73*, 1237.
4. (a) Tanaka, S.; Ichikawa, A. *J. Pharm. Sci.* **2006**, *101*, 19; (b) Huang, J.-F.; Thurmond, R. L. *Curr. Allergy Asthma Rep.* **2008**, *8*, 21.
5. http://www.clinicaltrials.gov/.
6. Lin, J.-S.; Dauvilliers, Y.; Arnulf, I.; Bastuji, H.; Anaclet, C.; Parmentier, R.; Kocher, L.; Yanagisawa, M.; Lehert, P.; Ligneau, X.; Perrin, D.; Robert, P.; Roux, M.; Lecomte, J.-M.; Schwartz, J.-C. *Neurobiol. Dis.* **2008**, *30*, 74.
7. (a) Ortiz de Montellano, P. R.; Almira Correia, M. In *Cytochrome P450, Structure, Mechanism and Biochemistry*; Ortiz de Montellano, P. R., Ed.; Plenum: New York, NY, 1995; p 307; (b) Yang, R.; Hey, J. A.; Aslanian, R.; Rizzo, C. A. *Pharmacology* **2002**, *66*, 128.
8. Pompni, S. A.; Gullo, V. P.; Horan, A. C.; Patel, M. G.; Coval, S. U.S. Patent 5,352,707, 1994.
9. Ganellin, C. R.; Leurquin, F.; Piripitsi, A.; Arrang, J.-M.; Garbarg, M.; Ligneau, X.; Schunack, W.; Schwartz, J.-C. *Arch. Pharm. Pharm. Med. Chem.* **1998**, *331*, 395.
10. Berlin, M.; Boyce, C. W. *Expert Opin. Ther. Patents* **2007**, *17*, 675.
11. Letavic, M. A.; Barbier, A. J.; Dvorak, C. A.; Carruthers, N. I.. In *Progress Med. Chem*; King, F. D., Lawton, G., Eds.; Elsevier Science, B.V.: Amsterdam, 2006; Vol. 14, p 181.
12. Zeng, Q; Aslanian, R. G.; Berlin, M.; Boyce, C.; Cao, J.; Mangiaracina, P.; McCormick, K. D.; Mutahi, M. Wa; Rosenblum, S. B.; Shih, N.-Y.; Solomon, D. S.; Ting, P. C.; Tom, W. U.S. Patent 7,105,505, 2006.
13. The syntheses of amines **8** are given as an example: to a 1,4-dioxane (30 mL) solution of **1** (3.0 g, 27.50 mmol) at 25 °C was added **2** (4.4 g, 27.50 mmol). The mixture was refluxed under N_2 for 48 h. After cooling to room temperature, the reaction was concentrated in vacuo and purified by 40 M Biotage cartridge to give **3** (64%). To a MeOH (50 mL) solution of **3** (4.4 g, 17.73 mmol) was added Ra-Ni (2.0 g) and the mixture was hydrogenated at 50 psi H_2 for 20 h. The reaction mixture was filtered through Celite, concentrated in vacuo, and purified by 40 M Biotage cartridge to give **4** (83%, $MH^+ = 219$). To a CH_2Cl_2 (50 mL) solution of **4** (3.2 g, 14.66 mmol) and picolinic acid (1.7 g, 14.66 mmol) were added DCC (3.9 g, 20.34 mmol) and HOBT (2.7 g, 20.34 mmol) at 25 °C. After stirring under N_2 for 20 h, water was added, the reaction was extracted with CH_2Cl_2 (2×), combined, washed with brine, dried over Na_2SO_4, filtered, concentrated in vacuo, and purified by 40 M Biotage cartridge to give **5** (48% $MH^+ = 324$). Compound **5** (2.1 g, 6.50 mmol) in 15 mL of acetic acid was heated at 120 °C under N_2 for 20 h. After cooling to room temperature, the reaction was concentrated in vacuo to give **6** (100%, $MH^+ = 306$). To an acetone (20 mL) solution of **6** (1.9 g, 6.22 mmol) was added K_2CO_3 (4.5 g, 32.48 mmol) at 25 °C. After stirring under N_2 for 40 min, 1-bromo-3-chloropropane (1.3 mL, 12.99 mmol) was added, and the mixture was refluxed for 20 h. After cooling to room temperature, the reaction mixture was filtered, concentrated in vacuo, and purified by 40 M Biotage cartridge to give **7** (93%, $MH^+ = 382$). To 1-mL glass tubes were added compound **7** (0.01 g, 0.026 mmol), MeCN (0.5 mL), and diisopropylethylamine (0.104 mmol). One molar stock solutions of each of the individual amines (0.053 mL, 0.053 mmol) were added to the tubes, which were then sealed and heated at 80 °C for 3 days. After cooling to room temperature, the solutions were transferred into the wells of a deep well polypropylene microtiter plate containing polystyrene isocyanate resin (2.5 equiv, 0.066 mmol) and MP-carbonate resin (4 equiv, 0.106 mmol). The microtiter plate was then sealed and shaken at 25 °C for 16 h. The solutions were filtered through a polypropylene frit into a collection plate. The wells of the top plate were then washed with MeCN (0.5 mL), and the plate was removed. After an aliquot of each solution was removed for LC/MS analysis, the remaining solutions in the collection plate were transferred into vials and the solvents removed in vacuo via a SpeedVac to provide amines **8**. Yield: 12–98%; purity >80% by LC/MS. Compounds **8b** and **8n**–**q** were obtained as mixtures of isomers.
14. For binding assays, membranes (P2 pellet) from rHu H_3–HEK cells (3 μg protein) were incubated in 200 μl of 50 mM Tris–HCl, pH 7.4, with 1 nM [^{3}H]N-α-methylhistamine (82 Ci/mmol) and compounds at concentrations equivalent to half orders of magnitude over a five-order of magnitude range. Nonspecific binding was determined in the presence of 10^{-5} M thioperamide. After 30 min at 30 °C, assay mixtures were filtered through 0.3% polyethylenimine-soaked GF/B glass fiber filters, which were rinsed thrice with buffer, dried, impregnated with Meltilex wax scintillant, and counted. K_i values were determined from curves fit to the data using GraphPad Prism nonlinear, least-squares, curve-fitting program.
15. Tang, W.; Kang, J.; Wu, X.; Rampe, D.; Wang, L.; Shen, H.; Li, Z.; Dunnington, D.; Garyantes, T. *J. Biomol. Screen.* **2001**, *6*, 325. Briefly, cells are loaded with rubidium for 3 h, then pre-equilibrated with drug for 30 min with Rb present. Extracellular Rb is washed out and KCl is used to depolarize and open the hERG channel to permit Rb^+ efflux. The supernatant is collected and Rb^+ is measured by flame atomic absorbance spectroscopy.
16. HEK293 cells expressing recombinant human histamine H_3-receptor were stimulated 30 min with 3 μM forskolin and compounds were characterized for their ability to reverse the inhibition of cAMP formation caused by 10^{-5} M *N*-α-methylhistamine over this time. cAMP assays were performed with an AlphaScreen cAMP assay kit.
17. Favreau, L.; Palamanda, J.; Lin, C.; Nomeir, A. *Drug Metab. Dispos.* **1999**, *27*, 436. and references therein.
18. Mei, H.; Korfmacher, W.; Morrison, R. *AAPS J.* **2006**, *8*, E493.