Pergamon

Bioorganic & Medicinal Chemistry Letters 12 (2002) 2035–2037

BIOORGANIC & MEDICINAL CHEMISTRY LETTERS

# Structure–Activity Relationships of Non-imidazole $H_3$ Receptor Ligands. Part 2: Binding Preference for D-Amino Acids Motifs

Ramin Faghih,* Wesley Dwight, Larry Black, Huaqing Liu, Robert Gentles, Kathleen Phelan, Timothy A. Esbenshade, Lynne Ireland, Thomas R. Miller, Chae-Hee Kang, Kathy M. Krueger, Gerard B. Fox, Arthur A. Hancock and Youssef L. Bennani

*Neuroscience Research, Global Pharmaceutical Research and Development, Abbott Laboratories, Abbott Park, IL 60064-6123, USA*

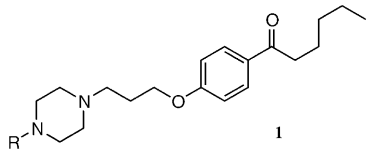
Received 2 November 2001; accepted 11 March 2002

**Abstract**—Structure–activity relationship studies on novel non-imidazole, D-amino acid containing ligands of histamine 3 receptors are presented. A-304121 is a D-alanine piperazine amide with high affinity at the rat $H_3$ receptor. 

Histamine mediates its action both in the CNS and the periphery through four distinct receptors ($H_1$–$H_4$) known to date.[1] Both agonists and antagonists of the histamine-3 receptor ($H_3R$) offer therapeutic potential since the receptor is involved in the presynaptic regulation of the release of not only histamine itself, but acetylcholine, gamma-aminobutyric acid, dopamine, noradrenaline and serotonin[2] as well. Potential therapeutic indications for novel $H_3$ ligands include cognitive disorders such as ADHD and Alzheimer's disease, or obesity.[3] In the preceding paper, we described efforts to delineate a pharmacophore for $H_3$ receptor blockade starting from a high throughput screening lead A-923, a non-imidazole lipophilic arylketone.[4] Early studies revealed the following: (1) the hydrophobic ketone region can be expanded somewhat both in size and substitution, (2) 'small-size' *N*-acyl substitutions with a basic site are tolerated and preferred, and (3) the piperazine basic site in A-923 is mandatory for binding/recognition of the ligand at the rat $H_3R$. In order to add the desired physicochemical features to improve drug-like characteristics, we explored this system further and introduced amino-acid attachments with the goal of increasing basicity towards a better balance of basicity/lipophilicity. We wish to report herein the SAR leading to the discovery of A-304121, a D-alanine derivative, which is a highly selective, potent ligand for the rat $H_3$ receptor.

Ketone **1** (R = Boc; Fig. 1) was prepared as described in the preceding paper (Scheme 1): the *N*-Boc group was removed under $TFA/CH_2Cl_2$ conditions. The free piperazine underwent *N*-Boc-amino-acid coupling using EDCI/DIEA/DMAP conditions to give compounds of the general structure in Figure 1. Table 1 displays binding results on human $H_1$, $H_2$, and rat cortical $H_3R$ for a select number of *N*-Boc and free amino-acid derivatives. These data are consistent with a trend of enhanced receptor affinity for *N*-Boc-D-amino acid containing compounds versus their natural (L) counterparts by a ratio of ~4–5. This trend was less pronounced when an additional basic site on the molecule was present (i.e., **8**/**9** and **16**/**17**; free amino amides). The enhanced affinity was indicative of the potential to find potent and selective compounds. From our studies, we found the corresponding cyclopropyl ketone to be well tolerated both in radioligand binding and in PK.

Intermediate **22** was prepared by treating commercially available chloro-4′-hydroxybutyrophenone **18** with concentrated NaOH and heat to induce cyclopropanation

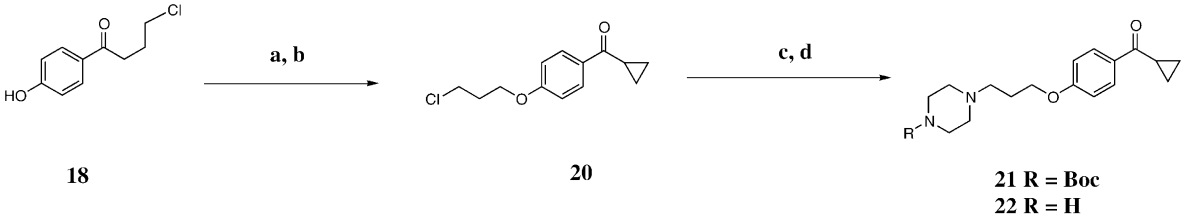



1

**Figure 1.**

*Corresponding author. Fax: +1-847-937-4143; e-mail: ramin.faghih@abbott.com

0960-894X/02/$ - see front matter 
PII: S0960-894X(02)00310-4




**Scheme 1.** (a) NaOH, heat, **19**; (b) Cl–$(CH_2)_3$–Br, $K_2CO_3$, 2-butanone, reflux 24 h; (c) *N*-Boc-piperazine, $KI/K_2CO_3$, 2-butanone-reflux 72 h; (d) TFA, $CH_2Cl_2$, 0 °C to rt, 12 h.

followed by *O*-alkylation with 1-bromo-3-chloropropane to give **20** (65% yield for two steps). Further displacement with *N*-Boc-piperazine and removal of the protecting group gave piperazine derivative **22**. Alternatively, piperazine **22** can be prepared directly from chloride **20** and 3 mol/equivalents of piperazine under $KI/K_2CO_3$ conditions.

Further acylations with various *N*-Boc-amino-acids, according to standard coupling procedures (see above) gave the corresponding desired products. TFA treatment produced the unprotected amino-acid derivatives, which were characterized and tested as either TFA or L-tartaric acid salts. A representative subset of compounds is shown in Table 2, along with their affinities at $H_1$, $H_2$ and $H_3$ receptors.

Data from Table 2 highlights several pharmacophore features: (1) Preference for D-amino acid (AA's) substituents is clear for most examples (e.g., **24** vs **25**; **53** vs **54**); (2) small AA's enhance receptor affinity (e.g., **24**, **25**, **43**, **47**, **55**, and **57**), whereas large aromatic groups (e.g., **31**, **34**, **35**, and **38**) are poorly tolerated; exception can be made for groups with increased chain flexibility (e.g., **29**) as well as groups containing a basic site (**33**), (3) β-alanine (e.g., **55**) provides the highest affinity ligand at rat $H_3$ receptors in this series; (4) chain substitutions (e.g., **57**–**60**) do not enhance binding affinity

**Table 1.** Binding affinities[a] ($pK_i$) at rat cortical $H_3$ receptors and human $H_1$ and $H_2$ receptors[5]

| Compd[b]: R | $H_3$ | $H_1$ | $H_2$ | Compd: R | $H_3$ | $H_1$ | $H_2$ |
|---|---|---|---|---|---|---|---|
| **2**: *N*-Boc-D-Ala | 8.91 | 5.17 | 4.97 | **10**: D-Ala | 8.72 | 4.45 | 4.39 |
| **3**: *N*-Boc-D-Ser | 8.62 | 4.77 | 4.64 | **11**: D-Ser | 8.15 | 4.33 | 4.46 |
| **4**: *N*-Boc-D-Val | 7.83 | 5.48 | 4.87 | **12**: D-Val | 7.77 | 4.78 | 4.50 |
| **5**: *N*-Boc-L-Val | 7.01 | 4.97 | 4.90 | **13**: L-Val | 7.10 | 4.72 | 4.42 |
| **6**: *N*-Boc-D-Leu | 7.84 | 5.33 | 5.35 | **14**: D-Leu | 7.21 | 4.84 | 4.85 |
| **7**: *N*-Boc-L-Leu | 6.95 | 5.09 | 5.46 | **15**: L-Leu | 6.77 | 5.01 | 4.78 |
| **8**: *N*-Boc-D-4-Pyridyl-Ala | 7.63 | 4.77 | 4.92 | **16**: D-4-Pyridyl-Ala | 8.04 | 5.48 | 4.43 |
| **9**: *N*-Boc-L-4-Pyridyl-Ala | 7.27 | 5.03 | 4.83 | **17**: L-4-Pyridyl-Ala | 7.99 | 5.64 | 4.46 |

[a]Values were estimated from at least three separated competition experiments (SEM $\leq 0.2$).
[b]Satisfactory $^1H$ NMR, MS spectra and elemental analyses were obtain for all new compounds.

**Table 2.** Binding affinities[a] ($pK_i$) at rat cortical $H_3$ receptors and human $H_1$ and $H_2$ receptors

| Compd: R | $H_3$ | $H_1$ | $H_2$ | Compd: R | $H_3$ | $H_1$ | $H_2$ |
|---|---|---|---|---|---|---|---|
| **23**: *N*-Boc-D-Ala | 8.68 | 5.09 | 4.08 | **42**: D-β-*tert*-Butyl-Ala | 7.28 | 4.66 | 4.72 |
| **24**: D-Ala | 8.90 | 4.16 | 4.15 | **43**: D-2-Abu | 8.80 | 4.15 | 4.00 |
| **25**: L-Ala | 7.69 | 4.27 | 4.38 | **44**: D-*tert*-Leu | 7.92 | 4.99 | 4.24 |
| **26**: *N*-Boc-D-Pro | 6.92 | 4.33 | 4.30 | **45**: D-Nva | 8.40 | 4.33 | 4.22 |
| **27**: D-Pro | 7.52 | 4.43 | 4.27 | **46**: D-β-Cyclohexyl-Ala | 6.79 | 4.84 | 4.69 |
| **28**: *N*-Boc-D-Phe | 8.40 | 5.26 | 5.00 | **47**: Sarcosine | 8.64 | 4.00 | 4.10 |
| **29**: D-HPhe | 8.34 | 5.00 | 5.00 | **48**: *N*-Isopropyl-D-Ala | 8.06 | 4.33 | 4.30 |
| **30**: D-Thi | 7.64 | 5.01 | 4.33 | **49**: *N*-Methyl-D-Phe | 7.36 | 4.87 | 4.75 |
| **31**: D-4-F-Phe | 7.53 | 5.03 | 4.40 | **50**: *N*-Methyl-D-Val | 7.12 | 4.46 | 4.31 |
| **32**: D-Thiazole-Ala | 8.24 | 4.00 | 4.01 | **51**: (*R*)-4-Oxo-2-Azetidine | 8.36 | 5.00 | 5.00 |
| **33**: D-His | 8.34 | 4.02 | 4.05 | **52**: (*S*)-4-Oxo-2-Azetidine | 7.37 | 4.66 | 4.57 |
| **34**: D-3-(1-Naphthyl)-Ala | 6.67 | 5.70 | 4.68 | **53**: D-Azetidine-2-CO | 8.88 | 4.90 | 4.85 |
| **35**: D-3-Benzothienyl-Ala | 6.54 | 5.36 | 4.82 | **54**: L-Azetidine-2-CO | 7.89 | 5.00 | 4.91 |
| **36**: D-Try | 7.52 | 5.01 | 4.48 | **55**: β-Ala | 9.30 | 4.55 | 4.00 |
| **37**: D-Phg | 7.56 | 5.34 | 4.61 | **56**: D-β-Hpro | 8.33 | 4.02 | 4.05 |
| **38**: D-Biphenyl-Ala | 6.88 | 4.92 | 4.69 | **57**: D-Abu | 8.71 | 5.68 | 4.96 |
| **39**: D-(2-INDA)-Gly | 7.58 | 4.97 | 4.72 | **58**: L-Abu | 8.60 | 5.33 | 4.97 |
| **40**: D-Cyano-Ala | 7.54 | 4.34 | 4.14 | **59**: D-α-Methyl-β-Ala | 8.19 | 5.82 | 4.99 |
| **41**: D-*O*-*tert*-Butyl-Ser | 8.07 | 4.79 | 4.34 | **60**: L-α-Methyl-β-Ala | 8.05 | 5.76 | 4.97 |

[a]See corresponding footnotes in Table 1.

(note that these latter enantiomeric derivatives have minimal affinity advantage over one another).

From the above studies, we selected A-304121 and A-308830 (e.g., **24** and **55**) based on their high affinity and selectivity for the rat $H_3$ receptor. D-Alanine derivative **24** and β-alanine **55** were tested at a number of biogenic amine receptors and transporters for the neurotransmitters norepinephrine, dopamine, and serotonin including different subtypes. Compounds **24** and **55** demonstrated low affinity at all of these sites generally with $pK_i < 5.00$. In order to test the hypothesis behind these studies, we evaluated the bioavailability of compounds **24** and **55** in rat after a dose of 5 mg/kg po. A-304121 showed an excellent oral bioavailability of 83% compared with 46 and 0%, respectively, for A-308830 (**55**) and A-923. Based on its bioavailability, potency and selectivity compound **24** (A-304121) was selected for further evaluation in models of in vitro functional activity. In the guinea pig isolated ileum assay[6] in which histamine $H_3$ receptors mediate inhibition of neurogenic contractions, A-304121 demonstrated a $pA_2$ value of 6.98. Compound **24** was also tested for $H_3$ receptor antagonist activity in an assay with $K^+$-evoked depolarization- induced release of [$^3$H]-histamine from rat synaptosomes[7] with $pK_b$ of 8.75.

In summary, we have discovered highly potent $H_3$ receptor antagonists. Additionally A-304121 (**24**) and compound **55** were shown to be more than 1000-fold selective for the $H_3$ receptor subtype over the $H_1$ and $H_2$ receptor subtypes. Thus, A-304121 and several analogues represent highly potent antagonists of the $H_3$ receptor, with potential clinical utility for treating CNS-related diseases.

## References and Notes


1. Hough, B. L. *Mol. Pharmacol.* **2001**, *59*, 415.
2. (a) Hill, S. J.; Gannelin, C. R.; Timmerman, H.; Schwartz, J. C.; Shankley, N. P.; Young, J. M.; Schunack, W.; Levi, R.; Haas, H. L. *Pharmacol. Rev.* **1997**, *49*, 253. (b) Arrang, J. M.; Garbarg, M.; Schwartz, J. C. *Nature* **1983**, *302*, 832. (c) Leurs, R.; Blandina, P.; Tedford, C.; Timmerman, H. *Trends Pharmacol. Sci.* **1998**, *19*, 177. (d) Lovenberg, T. W.; Roland, B. L.; Wilson, S. J.; Jiang, X.; Pyati, J.; Huvar, A.; Jackson, M. R.; Erlander, M. G. *Mol. Pharmacol.* **1999**, *55*, 1101.
3. (a) *The Histamine $H_3$-Receptor: A Target for New Drugs*. Leurs, R., Timmerman, H., Eds. Elsevier: Amersterdam, 1998. (b) Yates, S. L.; Pawlowski, G. P.; Antal, J. M.; Ali, S. M.; Jiang, J.; Brunden, K. R. *Soc. Neurosci Abstr.* **2000**, *26*, 102. 10. (c) Ligneau, X.; Lin, J.-S.; Vanni-Mercier, G.; Jouvet, M.; Muir, J. L.; Ganellin, C. R.; Stark, H.; Elz, S.; Schunack, W.; Schwartz, J.-C. *J. Pharmacol. Exp. Ther.* **1998**, *287*, 658. (d) Passini, M. B.; Bacciottini, L.; Mannaioni, P. F.; Blandina, P. *Neurosci. Biobehav. Rev.* **2000**, *24*, 107. (e) Ali, S. M.; Tedford, C. E.; Gregory, R.; Handley, M. K.; Yates, S. L.; Hirth, W. W.; Phillips, J. G. *J. Med. Chem.* **1999**, *42*, 903. (f) Stark, H.; Sadek, B.; Krausse, M.; Huls, A.; Ligneau, X.; Ganellin, C. R.; Arrang, J.-M.; Schwartz, J. C.; Schunack, W. *J. Med. Chem.* **2000**, *43*, 3987.
4. Faghih, R.; Dwight, W.; Gentles, R.; Phelan, K.; Esbenshade, T.; Ireland, L.; Miller, T.; Kang, C.-H.; Krueger, K.; Hancock, A. A.; Bennani; Y. L. *Bioorg. Med. Chem. Lett.* (see preceding paper).
5. Hancock, A. A.; Esbenshade, T. E. In *Current Protocols In Pharmacology*; Enna, S., Williams, M., Ferkany, J., Kenakin, T., Porsolt, R., Sullivan, J., Eds.; John Wiley & Sons: New York, 2000; Vol. 1.19, p. 1.
6. Ireland-Denny, L.; Parihar, A. S.; Miller, T. R.; Kang, C.-H.; Krueger, K. M.; Esbenshade, T. A.; Hancock, A. A. *Eur. J. Pharmacol.* **2001**, *433*, 141.
7. Garbarg, M.; Arrang, J.-M.; Rouleau, A.; Ligneau, X.; Trung Tuong, M. D.; Schwartz, J.-C.; Ganellin, C. R. *J. Pharmacol. Exp. Ther.* **1992**, *263*, 304.