A New Potent and Selective Histamine H_3 Receptor Agonist, 4-(1*H*-Imidazol-4-ylmethyl)piperidine

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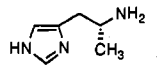
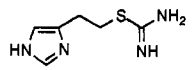
It has been shown that the presynaptic histamine H_3 receptor[1] regulates not only the release and synthesis of histamine, but also the release of other neurotransmitters[2,3] and can be regarded as a potential target for new therapeutics.[4,5] Until now only a few potent and selective agonists for the histamine H_3 receptor have been described. Methylation of the side chain of histamine has resulted in agonists like N^{α}-methylhistamine[1] and the chiral agonists (*R*)-α-methylhistamine[6] (1) and α(*R*),β(*S*)-dimethylhistamine.[7,8] Out of this series of methylated histamine analogues, the (*R*)-enantiomer of α-methylhistamine 1 has been used extensively as a pharmacological tool. Recently the nonchiral histamine H_3 agonist, imetit (2), has been described.[9-12] This agonist is different from histamine and its methylated analogues because it has a planar basic isothiourea group instead of an amino group. Imetit (2) and (*R*)-α-methylhistamine (1) are equipotent on the H_3 receptor as reported by Van der Goot *et al.* and Howson *et al.* on the inhibition of the electrically evoked twitches of the guinea pig ileum (jejunum).[9,10]

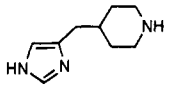


1, (R)-α-methylhistamine



2, imetit

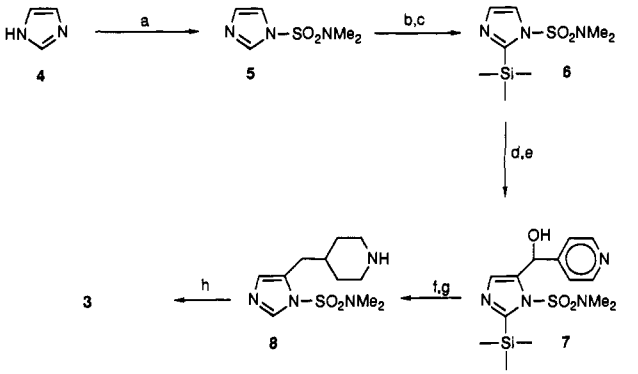
We now describe a new, potent and selective nonchiral histamine H_3 agonist, 4-(1*H*-imidazol-4-ylmethyl)piperidine (immepip, 3) as prepared from a series of histamine analogues[13] in which we incorporated the amino group in various ring structures in order to obtain more information about the influence and the optimal location of the amino group relative to the imidazole ring. For 3 the alkyl side chain was extended to a length of four methylene groups, and the amino group was incorporated in a piperidine ring.



3, immepip

Compound 3 was synthesized by the direct coupling of 4-pyridinecarboxaldehyde to the 5-position of a suitable 1,2-diprotected imidazole 6 by lithiation (Scheme 1).[14] The hydroxyl group of 7 was removed by acylation and subsequent hydrogenation at 50 atm using Pd/C as a catalyst. Under these conditions the pyridine ring is also reduced to a piperidine ring and the trimethylsilyl protecting group is hydrolyzed. After removal of the *N*,*N*-dimethylsulfonamide protecting group, immepip (3) was isolated as the dihydrobromide (overall yield was 20%).

Scheme 1[a]



[a] Reagents used: (a) *N*,*N*-dimethylsulfamoyl chloride, Et_3N, toluene; (b) *n*-BuLi, THF, −70 °C; (c) trimethylsilyl chloride; (d) *n*-BuLi, THF, −70 °C; (e) 4-pyridinecarboxaldehyde; (f) DBU, Ac_2O; (g) H_2, Pd/C, 50 atm; (h) 30% HBr, reflux.

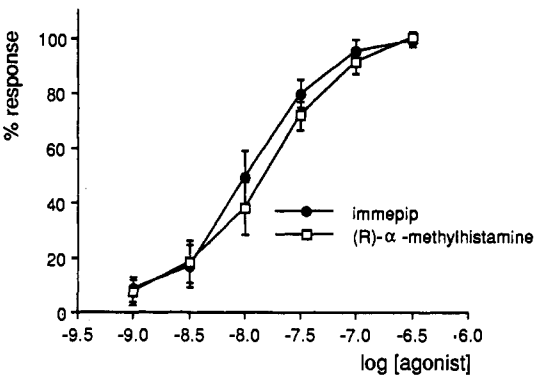


Figure 1. Concentration response curves for immepip (n = 8) and (*R*)-α-methylhistamine (n = 22), constructed from the inhibition of the electrically evoked twitches of the guinea pig jejunum.[15] n represents the number of animals used. Values shown in the graph are expressed as mean ± sd.

The H_3 activity was functionally determined on an *in vitro* test system based on the concentration-dependent inhibition of electrically evoked twitches of isolated guinea pig jejunum segments by histamine H_3 agonists.[15] Average concentration response curves (CRC's) for immepip (3) and (*R*)-α-methylhistamine (1) (for comparison) are shown in Figure 1. From this graph it is clear that 3 is equipotent as (or even slightly more active than) (*R*)-α-methylhistamine (1) on the H_3 receptor. The pD_2 value for 3 as determined on jejunum preparations of eight different animals was 8.0 ± 0.1 (mean ± sd). For comparison, (*R*)-α-methylhistamine (1) has a pD_2 value of 7.8 ± 0.2 (n = 22) on this test system. The H_3 antagonist thioperamide caused a rightward parallel shift of the CRC for immepip (3). The pA_2 value of thioperamide using 3 as an agonist, was 8.2 ± 0.2 with a Schild slope of 0.8 ± 0.1 (n = 3) (not significantly different from unity). This is slightly lower than the pA_2 value of thioperamide, obtained using (*R*)-α-methylhistamine (1) on this assay.[15] This lower affinity has also been reported, using imetit (2) as agonist.[12] The potent agonistic activity of 3 on the H_3 receptor was confirmed in radioligand binding studies (Figure 2).

Displacement of the H_3 antagonist [^{125}I]iodophenpropit[16,17] binding to rat cortex membranes resulted in

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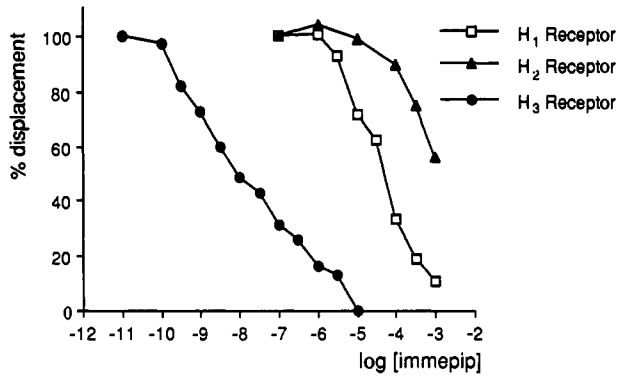

Figure 2. Receptor selectivity of immepip as measured by radioligand binding studies. Displacement of the specific binding ligands in representative experiments is shown. Competition of immepip with [^{3}H]mepyramine binding to membranes of CHO cells expressing guinea pig H_1 receptors,[19] [^{125}I]iodoaminopotentidine binding to membranes of CHO cells expressing human H_2 receptors[20,24] and [^{125}I]iodophenpropit binding to membranes from rat cortex[17] was measured in at least three independent experiments, performed in triplicate.

shallow displacement curves for immepip (**3**). Computer analysis of these data reveals two binding sites (using the program LIGAND.[18] This is in agreement with the described displacement curves for other H_3 agonists and indicative for the interaction of the H_3 receptor with a G-protein. The K_H and the K_L for **3** on the H_3 receptor are 2.7 ± 0.5 nM and 1.01 ± 0.2 μM, respectively. For comparison, (*R*)-α-methylhistamine (**1**) showed a K_H and a K_L of 4.3 ± 3.4 nM and 0.22 ± 0.15 μM, respectively, on the same assay.[17] From Figure 2 it is also clear that immepip (**3**) is highly selective for the H_3 receptor. The pK_i of **3** for the guinea pig H_1 receptor was 4.79 ± 0.10 (using [^{3}H]mepyramine as a radioligand[19]), whereas its affinity for the human H_2 receptor[20] was too low to be determined accurately (using [^{125}I]iodoaminopotentidine as a radioligand; $pK_i < 3.5$).

If we compare the structure of the methylated histamine analogues, imetit and immepip, some interesting observations can be made. The amino group of histamine and its methylated analogues is protonated at physiological pH[21] and is located at a distance of two methylene groups (≈4.5 Å) away from the imidazole ring. This ammonium group could interact with a carboxylate group in the receptor, as postulated for the H_2 receptor.[22,23] The isothiourea group of imetit (**2**) is also protonated at a pH of 7.4.[11] This means that the isothiouronium group can also interact with a carboxylate group. However, since only the imino nitrogen of the isothiourea group can be protonated, the distance between the imidazole ring and the hydrogen donating nitrogens is not two methylene groups (≈4.5 Å) as in histamine and its methylated analogues, but longer (≈8 Å). For **3** it is obvious that the proton-donating ammonium group is located at a distance of four methylene groups from the imidazole ring (≈7.5 Å). These observations make immepip (**3**), together with imetit (**2**) and the methylated analogues a valuable tool in molecular modeling studies.

It can be concluded that immepip (**3**) is a new and selective histamine H_3 agonist, equipotent as (*R*)-α-methylhistamine (**1**) and imetit (**2**), which can be useful as a pharmacological tool and perhaps as a therapeutical agent, but also, because of its distinctive structure, for SAR and molecular modeling studies.

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References

(1) Arrang, J. M.; Garbarg, M.; Schwartz, J. C. Auto-inhibition of brain histamine release mediated by a novel class (H_3) of histamine receptor. *Nature* **1983**, *302*, 832–837.
(2) Leurs, R.; Timmerman, H. The histamine H_3-receptor: A target for developing new drugs. *Prog. Drug Res.* **1992**, *39*, 127–165.
(3) Lipp, R.; Stark, H.; Schunack, W. Pharmacochemistry of H_3-Receptors: The Histamine Receptor. In *Receptor Biochemistry and Methodology*, 16th ed.; Schwartz, J.-C., Haas, H. L., Eds.; Wiley-Liss, Inc.: New York, 1992; pp 57–72.
(4) Timmerman, H. Histamine H_3-ligands: just pharmacological tools or potential therapeutic agents? *J. Med. Chem.* **1990**, *33*, 4–11.
(5) Schwartz, J. C.; Arrang, J. M.; Garbarg, M.; Pollard, H. A third histamine receptor subtype: characterisation, localisation and functions of the H_3-receptor. *Agents Actions* **1990**, *30*, 13–23.
(6) Arrang, J. M.; Garbarg, M.; Lancelot, J. C.; Lecomte, J. M.; Pollard, H.; Robba, M.; Schunack, W.; Schwartz, J. C. Highly potent and selective ligands for histamine H_3-receptors. *Nature* **1987**, *327*, 117–123.
(7) Lipp, R.; Arrang, J. M.; Buschmann, J.; Garbarg, M.; Luger, P.; Schunack, W.; Schwartz, J. C. Novel chiral H_3-receptor agonists. In *New perspectives in Histamine Research*; Birkhäuser Verlag: Basel, 1991; pp 277–282.
(8) Lipp, R.; Arrang, J. M.; Garbarg, M.; Luger, P.; Schwartz, J. C.; Schunack, W. Synthesis, Absolute Configuration, Stereoselectivity, and Receptor Selectivity of (αR,βS)-α,β-Dimethylhistamine, a Novel Highly Potent Histamine H_3 Receptor Agonist. *J. Med. Chem.* **1992**, *35*, 4434–4441.
(9) Van der Goot, H.; Schepers, M. J. P.; Sterk, G. J.; Timmerman, H. Isothiourea analogues of histamine as potent agonists or antagonists of the histamine H_3-receptor. *Eur. J. Med. Chem.* **1992**, *27*, 511–517.
(10) Howson, W.; Parsons, M. E.; Raval, P.; Swayne, G. T. G. Two novel, potent and selective histamine H_3 receptor agonists. *Bioorg. Med. Chem. Lett.* **1992**, *2*, 77–78.
(11) Ganellin, C. R.; Bangandersen, B.; Khalaf, Y. S.; Tertiuk, W.; Arrang, J. M.; Garbarg, M.; Ligneau, X.; Rouleau, A.; Schwartz, J. C. Imetit and N-Methyl Derivatives. The Transition from Potent Agonist to Antagonist at Histamine H_3 Receptors. *Bioorg. Med. Chem. Lett.* **1992**, *2*, 1231–1234.
(12) Garbarg, M.; Arrang, J. M.; Rouleau, A.; Ligneau, X.; Tuong, M. D. T.; Schwartz, J. C.; Ganellin, C. R. S-[2-(4-Imidazolyl)ethyl]-isothiourea, a Highly Specific and Potent Histamine H_3 Receptor Agonist. *J. Pharmacol. Exp. Ther.* **1992**, *263*, 304–310.
(13) Vollinga *et al.* Unpublished results.
(14) Vollinga, R. C.; Menge, W. M. P. B.; Timmerman, H. A New Convenient Route for the Synthesis of 4(5)-(ω-Aminoalkyl)-1H-Imidazoles. *Recl. Trav. Chim. Pays-Bas* **1993**, *112*, 123–125.
(15) Vollinga, R. C.; Zuiderveld, O. P.; Scheerens, H.; Bast, A.; Timmerman, H. A Simple and Rapid *In vitro* Test System for the Screening of Histamine H_3 Ligands. *Meth. Find. Exp. Clin. Pharmacol.* **1992**, *14*, 747–751.
(16) Menge, W. M. P. B.; Van der Goot, H.; Timmerman, H.; Eersels, J. L. H.; Herscheid, J. D. M. Synthesis of S-[3-(4(5)-Imidazolyl)-Propyl],N-[2-(4-[^{125}I])-Iodophenyl)Ethyl]Isothiourea Sulfate (^{125}I-Iodophenpropit), a New Probe for Histamine H_3 Receptor Binding Sites. *J. Labelled Comp. Radiopharm.* **1992**, *31*, 781–786.
(17) Jansen, F. P.; Rademaker, B.; Bast, A.; Timmerman, H. The first Radiolabeled Histamine H_3 Receptor Antagonist, [^{125}I]iodophenpropit: Saturable and Reversible Binding to Rat Cortex Membranes. *Eur. J. Pharmacol.* **1992**, *217*, 203–205.
(18) Munson, P. J.; Rodbard, D. Ligand, a versatile computerized approach for characterization of ligand-binding systems. *Anal. Biochem.* **1980**, *107*, 220–239.
(19) Traiffort, E.; Leurs, R.; Arrang, J. M.; Tardivel-Lacombe, J.; Diaz, J.; Ruat, M.; Schwartz, J. C. Guinea-pig histamine H_1 receptor: Gene cloning, characterization and tissue expression revealed by in situ hybridization. *J. Neurochem.* In press.
(20) Gantz, I.; Munzert, G.; Tashiro, T.; Schäffer, M.; Wang, L.; Delvalle, J.; Yamada, T. Molecular cloning of the human histamine H_2 receptor *Biochem. Biophys. Res. Commun.* **1991**, *178*, 1386–1392.
(21) Ganellin, C. R. Medicinal Chemistry and dynamic structure-activity analysis in the discovery of drugs acting at histamine H_2 receptors. *J. Med. Chem.* **1981**, *24*, 913–920.
(22) Haaksma, E. E. J. Development of theoretical models for activation and recognition of the histamine H_2-receptor by H_2-receptor agonists and antagonists. Ph.D. Thesis, Vrije Universiteit, Amsterdam, 1991.
(23) Timmerman, H. Routes to Histamine H_2-Agonists. *Quant. Struct. Act. Rel.* **1992**, *11*, 219–223.
(24) Leurs, R. *et al.* Unpublished results.