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Synthesis and biological assays of new H_3-antagonists with imidazole and imidazoline polar groups

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Abstract

New histamine H_3-receptor antagonists were synthesised and tested on rat brain membranes and on electrically stimulated guinea-pig ileum. The new compounds have a central polar group represented by a 2-alkylimidazole or a 2-thioimidazoline nucleus. The effect of the polar group basicity on the optimal length of the alkyl chain, connecting this group to a 4(5)-imidazolyl ring, was investigated. The best affinity values, obtained by displacement of [^{3}H]-RAMHA from rat brain, were obtained for the 2-alkylimidazole derivatives (**2a**–**f**) with tetramethylene chain (pK_i 8.03–8.97), having an intermediate basicity between that of the previously reported 2-thioimidazoles (**1a**–**i**) and that of 2-alkylthioimidazolines (**3a**–**h**). In contrast, a general lowering of affinity (pK_i 5.90–7.63) was observed for compounds of the last series (**3a**–**h**), with a complex dependence on the terminal lipophilic group and chain length.

Keywords: Histamine; H_3-receptor antagonists; 2-Alkylimidazoles; 2-Thioimidazolines

1. Introduction

Histamine, besides its well-known actions on blood vessels and gastric secretion, is known to act as a neurotransmitter in the central nervous system (CNS) [1], exerting its pharmacological actions through the activation of distinct receptor subtypes, named H_1, H_2 and H_3. The histamine H_3-receptor [2] is located presynaptically on various kinds of central neurons, regulating the synthesis and the release of histamine [3,4] and of other neurotransmitters [5], such as noradrenaline [6], acetylcholine [7], dopamine [8] and serotonin [9,10]. Moreover, H_3-receptors can also play a modulating role in peripheral neurotransmission [11,12].

The central effects of the H_3-receptor antagonists suggest a potential therapeutic role for the treatment of some neurological disorders, such as narcolepsy, attention-deficit hyperactive disorder, epilepsy and obesity [13,14]. These possibilities prompted the search for new potent, selective and brain-penetrating H_3-receptor antagonists.

CNS access can be reduced for various reasons, such as a high binding to plasma proteins (e.g. for thioperamide [15,16]) or drug protonation (e.g. for clobenpropit); these phenomena lead to high peripheral concentrations compared to brain levels, which should be considered as being a drawback, given the possible actions of H_3-antagonists on vessels, lung and the gastrointestinal tract [17,18].

The basicity of the so-called 'polar group', connected to an imidazole headgroup by an alkyl spacer in the classical H_3-antagonist formula [19], is therefore a crucial feature for the pharmacological optimisation of these compounds. Indeed, a classification of H_3-antagonists based on the nature of the polar group can be proposed from a structural viewpoint, grouping them into basic (e.g. clobenpropit) and neutral (e.g. thioperamide) ones [20].

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Recently, a new series of H_3-antagonists has been described, having no traditional polar group, but a double or a triple bond between the alkyl chain and a lipophilic ending group [21]. While this casts doubts on the classical definition cited above, the defects of which are further underlined by the report of potent antagonists lacking the imidazole headgroup [22], the study of the possible interactions occurring at the polar group is still important for the design of new compounds, as it can provide information on a group that could be tightly bound to the receptor. Such a group could therefore be considered as a good starting point for further structural modifications, aimed at the optimisation of pharmacokinetic and toxicological properties.

In this field, we have tested different heterocyclic groups [23], directing our attention towards the importance of basicity in polar groups and its relation with the alkyl chain length. In fact, it is our opinion that SAR profiles for the two classes of H_3-antagonists cited are different and that, while strongly basic compounds should be avoided to ensure an easy crossing of the blood–brain barrier, polar interactions of basic groups (such as the isothiourea one in clobenpropit, or the guanidine one in thioperamide derivatives [24]) at the H_3-receptor could be exploited for the design of potent ligands. The rationale of the present study was to test the possibility of obtaining good affinity with heterocyclic polar groups having intermediate basicity, and thus of being able to take polar interaction at the receptor, while conserving a significant fraction of neutral species in solution at physiological pH, to allow passive penetration of lipophilic membranes.

Starting from a series of known H_3-antagonists with 2-alkylthioimidazole polar group (**1**) [20], which are not protonated at physiological pH ($pK_a \sim 4$, [25]), we synthesised and tested two new series of compounds (2-alkylimidazoles (**2**) and 2-alkylthioimidazolines (**3**)) which have a polar group with a higher basicity than the previous series **1**.

We also investigated the effect of the polar group basicity on the optimal length of the alkyl chain, preparing compounds with different spacer lengths. In fact, in the neutral 2-alkylthioimidazole derivatives (**1**), the best H_3-receptor antagonist activity and affinity values were obtained with the shortest chains (two-methylene spacer) [20], and this is in contrast with what has been reported for basic H_3-antagonists [26].

2. Pharmacology

Newly synthesised compounds were tested for their H_3-receptor affinity on rat brain membranes, by displacement of [^{3}H]-(R)-α-methylhistamine ([^{3}H]-RAMHA) binding to cerebral cortex synaptosomes.

Histamine H_3-receptor antagonist potency was evaluated on electrically stimulated guinea-pig ileum, by inhibition of RAMHA-induced responses [27].

3. Chemistry

4(5)Phenyl-2-[ω-[4(5)imidazolyl]alkyl]imidazoles **2a**, **2c**, **2e** were prepared starting from the corresponding 4(5)-(ω-cyanoalkyl)imidazoles, as described in Scheme 1. These were obtained by substitution with sodium cyanide from alkyl halides, prepared following different synthetic routes, depending on the length of the chain.

4(5)-(β-Chloroethyl)imidazole was prepared from histamine by diazotisation and substitution with $SOCl_2$, as described in a previous paper [28]; the homologue chloropropyl derivative was obtained in the same way from 4(5)-(γ-hydroxypropyl)imidazole, which was synthesised from 3,4-dihydro-2*H*-pyran by a series of reactions described in the literature [29,30].

4(5)-(δ-Cyanobutyl)imidazole was prepared from 2-(*t*-butyldimethylsilyl)-5-(4-chlorobutyl)-1-(dimethylsulfamoyl)imidazole, described by Vollinga et al. [31], by treatment with sodium cyanide of the intermediate partially deprotected in position 2, and following deprotection of position 1 by aqueous HCl.

All the cyano-compounds were then treated with absolute EtOH and gaseous HCl to give the iminoether derivatives which, by cyclisation with ω-bromoacetophenone, gave the final products.

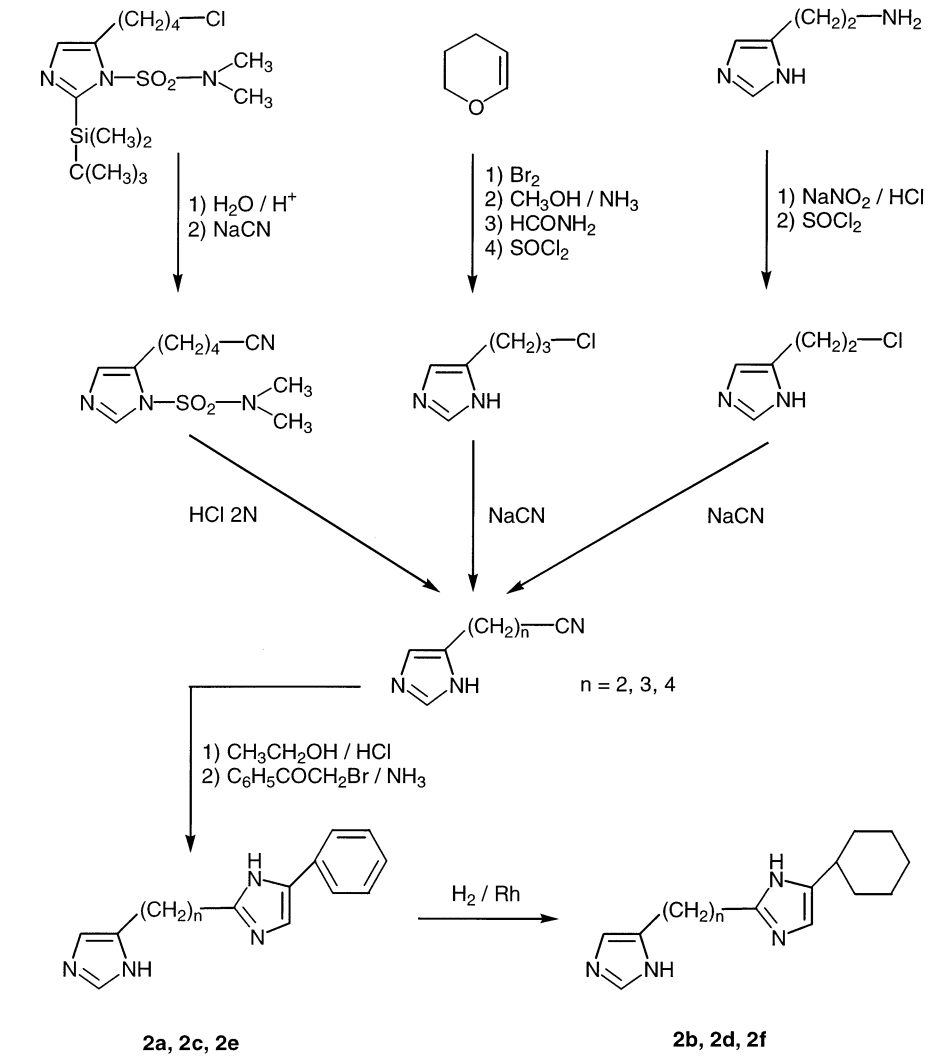
4(5)Cyclohexyl-2-[ω-[4(5)imidazolyl]alkyl]imidazoles **2b**, **2d**, **2f** were prepared by the hydrogenation of compounds **2a**, **2c**, **2e** with rhodium catalyst.

Imidazoline derivatives **3a**–**3h** were obtained by condensation of the appropriate 4(5)-(ω-chloroalkyl)-imidazole with substituted 2-thioimidazolines [32–34], obtained by cyclisation of the corresponding diamine with CS_2, as described in Scheme 2.

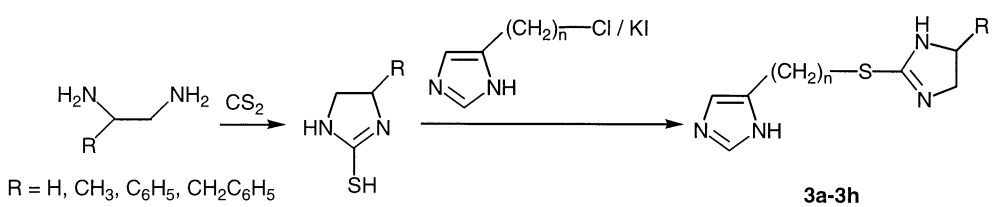
4. Experimental

4.1. Chemistry

Melting points were not corrected and were determined using a Büchi instrument (Tottoli). The newly synthesised substances were analysed for C, H and N. The percentages we found were within ± 0.4 of the theoretical values. The ^{1}H NMR spectra were recorded on a Bruker 300 spectrometer (300 MHz) with tetramethylsilane (TMS) as an internal standard. Mass spectra were recorded using a Finnigan MAT SSQ 710 instrument, and IR spectra were recorded using a Jasco FTIR 300E instrument. Reactions were followed by TLC, on a Kieselgel 60 F 254 (DC-Alufolien, Merck).



Scheme 1.



Scheme 2.

Final compounds and intermediates were purified by chromatography on a preparative Gilson HPLC, using an SiO_2 column (LiChroprep, Si 60, 15–25 μm, Merck). When indicated, the methanol phase was saturated with gaseous NH_3.

Yields and characteristic data of the final products are described in Table 1.

4.1.1. 4(5)-(β-cyanoethyl)imidazole and 4(5)-(γ-cyanopropyl)imidazole

Sodium cyanide (6.7 mmol) was added to a solution of 4.5 mmol of the appropriate 4(5)-(ω-chloroalkyl)-imidazole (prepared according to Refs. [28–30]) in the minimum amount of DMF. The reaction mixture was stirred at 70°C for 20 h, under dry nitrogen. The solvent was then evaporated under reduced pressure and the residual oil was purified by column chromatography (SiO_2, CH_2Cl_2–$CH_3OH(NH_3) = 20:1$), giving solid products.

M.p. 4(5)-(β-cyanoethyl)imidazole 96–99°C [35]; m.p. 4(5)-(γ-cyanopropyl)imidazole 132–134°C [35].

Table 1
Yields and characteristic data of final products

Comp.	Yield (%)	Crystallisation solvent	M.p. (°C) [a]	Analysis
2a	49	*i*-PrOH/Et_2O	234–236	$C_{14}H_{14}N_4 \cdot 2HCl$
2b	80	abs EtOH/Et_2O	175–176	$C_{14}H_{20}N_4 \cdot 2C_4H_4O_4$
2c	51	*i*-PrOH/Et_2O	253–254	$C_{15}H_{16}N_4 \cdot 2HCl$
2d	82	abs EtOH/Et_2O	156–159	$C_{15}H_{22}N_4 \cdot 2C_6H_3N_3O_7$
2e	47	abs EtOH/Et_2O	205–206	$C_{16}H_{18}N_4 \cdot 2HCl$
2f	79	abs EtOH	105–107	$C_{16}H_{24}N_4 \cdot 2C_2H_2O_4$
3a	46	abs EtOH	213–215	$C_8H_{12}N_4S \cdot 2HCl \cdot 0.5H_2O$
3b	58	MeOH/acetone	182–184	$C_9H_{14}N_4S \cdot 1.5C_2H_2O_4$
3c	46	abs EtOH/Et_2O	205–207	$C_{14}H_{16}N_4S \cdot HCl \cdot HI$
3d	42	abs EtOH/acetone	190–193	$C_{15}H_{18}N_4S \cdot 2HCl$
3e	38	abs EtOH	188–189	$C_9H_{14}N_4S \cdot 2HCl$
3f	18	abs EtOH	142–144	$C_{10}H_{16}N_4S \cdot 2C_2H_2O_4$
3g	30	abs EtOH	221–223	$C_{15}H_{18}N_4S \cdot 2C_{10}H_8N_4O_5$
3h	20	abs EtOH/acetone	135–136	$C_{16}H_{20}N_4S \cdot 2C_2H_2O_4$

[a] The melting points refer to the analysed salts.

4.1.2. 4(5)-(4-cyanobutyl)imidazole

2-(*t*-Butyldimethylsilyl)-1-(dimethylsulfamoyl)-5-(4-chlorobutyl)imidazole (8 mmol), prepared according to Ref. [31], was deprotected in position 2 by 1 N HCl and dissolved in the minimum amount of DMF with 24 mmol of sodium cyanide. The reaction mixture was stirred at 130°C for 4 h, under dry nitrogen. The solvent was then evaporated under reduced pressure and the semisolid residue was dissolved in water. Extraction with ether and evaporation of the dried organic phase gave 1-(dimethylsulfamoyl)-5-(4-cyanobutyl)-imidazole (yield 53%). The N-deprotection was achieved with 2 N HCl. The aqueous acid solution was then treated with K_2CO_3 and the product was extracted with CH_2Cl_2. The organic layer was dried with Na_2SO_4 and evaporated under reduced pressure to give 0.49 g of 4(5)-(4-cyanobutyl)imidazole (yield 71%).

^{1}H NMR (DMSO-d_6) (free base): δ 1.61 (m, 4H, CH_2–*CH_2*–*CH_2*–CH_2), 2.49 (t, 2H, CH_2), 2.52 (t, 2H, CH_2), 6.77 (s, 1H, Im-5-H), 7.56 (s, 1H, Im-2-H).

*4.1.3. General method of preparation of 4(5)-phenyl-2-[ω-[4(5)imidazolyl)alkyl]imidazoles (**2a**, **2c**, **2e**)*

Gaseous HCl (9 mmol) was added to a solution of 3 mmol of the appropriate 4(5)-(ω-cyanoalkyl)imidazole in 3.1 mmol of absolute EtOH, maintaining the temperature below 5°C. The resulting mixture was kept at 4°C for 5 days. ω-Bromoacetophenone (3.1 mmol) and gaseous NH_3 (2.6 g, 0.15 mol) were added to a solution of the crude product (dihydrochloride) in monoethylene glycol, and the mixture was cooled to −20°C. The solution was then heated under pressure at 110°C for about 30 h.

The compounds were purified by column chromatography (SiO_2, **2a**: CH_2Cl_2–$CH_3OH(NH_3)$ = 12:1; **2c** and **2e**: CH_2Cl_2–$CH_3OH(NH_3)$ = 10:1). Yields and characteristic data of these compounds are reported in Table 1.

^{1}H NMR (DMSO-d_6) **2a** (dihydrochloride): δ 3.31 (t, J = 6.9 Hz, 2H, CH_2), 3.43 (t, J = 6.9 Hz, 2H, CH_2), 7.41 (t, 1H, Ph-4-H), 7.49 (t, 2H, Ph-3-H), 7.52 (s, 1H, Im-5-H), 7.90 (d, 2H, Ph-2-H), 8.03 (s, 1H, Im-5-H), 9.02 (s, 1H, Im-2-H).

^{1}H NMR (DMSO-d_6) **2c** (free base): δ 1.91–2.00 (m, 2H, CH_2–*CH_2*–CH_2), 2.53 (t, 2H, CH_2), 2.66 (t, 2H, CH_2), 6.75 (s, 1H, Im-5-H), 7.15 (t, 1H, Ph-4-H), 7.31 (t, 2H, Ph-3-H), 7.40 (s, 1H, Im-5-H), 7.51 (s, 1H, Im-2-H), 7.70 (d, 2H, Ph-2-H).

^{1}H NMR (DMSO-d_6) **2e** (free base): δ 1.58–1.72 (m, 4H, CH_2–*CH_2*–*CH_2*–CH_2), 2.52 (t, 2H, CH_2), 2.65 (t, 2H, CH_2), 6.71 (s, 1H, Im-5-H), 7.14 (t, 1H, Ph-4-H), 7.31 (t, 2H, Ph-3-H), 7.38 (s, 1H, Im-5-H), 7.49 (s, 1H, Im-2-H), 7.68 (d, 2H, Ph-2-H).

*4.1.4. General method of preparation of 4(5)cyclohexyl-2-[ω-[4(5)imidazolyl)alkyl]imidazoles (**2b**, **2d**, **2f**)*

A solution of compounds **2a**, **2c** or **2e** (1.1 mmol) in 35 ml of 3 N HCl was hydrogenated using a Parr apparatus with 0.5 g of Rh/C (5%) at room temperature, for 65 h. The products obtained after filtration of the catalyst were purified by column chromatography (SiO_2, **2b**: CH_2Cl_2–$CH_3OH(NH_3)$ = 7:2; **2d**: CH_2Cl_2–$CH_3OH(NH_3)$ = 12:1; **2f**: CH_2Cl_2–$CH_3OH(NH_3)$ = 9:1). Yields and characteristic data of these compounds are reported in Table 1.

^{1}H NMR (DMSO-d_6) **2b** (difumarate): δ 1.20–1.38 (m, 6H, C_6H_{11}), 1.63–1.72 (m, 5H, C_6H_{11}), 2.92 (t, J = 6.9 Hz, 2H, CH_2), 3.03 (t, J = 6.9 Hz, 2H, CH_2), 6.58 (s, 1H, Im-5-H), 6.79 (s, 1H, Im-5-H), 7.66 (s, 1H, Im-2-H).

^{1}H NMR (DMSO-d_6) **2d** (free base): δ 1.23–1.29 (m, 6H, C_6H_{11}), 1.62–1.89 (m, 7H, CH_2–*CH_2*–CH_2–CH_2–CH_2–CH_2; C_6H_{11}), 1.86 (t, 2H, CH_2), 2.51 (t, 2H, CH_2), 6.47 (s, 1H, Im-5-H), 6.72 (s, 1H, Im-5-H), 7.48 (s, 1H, Im-2-H).

^{1}H NMR (DMSO-d_6) **2f** (free base): δ 1.17–1.28 (m, 6H, C_6H_{11}), 1.56–1.69 (m, 9H, CH_2–CH_2–CH_2–CH_2; C_6H_{11}), 2.49 (t, 2H, CH_2), 2.57 (t, 2H, CH_2), 6.52 (s, 1H, Im-5-H), 6.69 (s, 1H, Im-5-H), 7.50 (s, 1H, Im-2-H).

4.1.5. General method of cyclisation of the diamines with CS_2

A solution of 0.125 mol of the appropriate diamine in 30 ml of EtOH was added to a solution of 32 ml of CS_2 and 32 ml of EtOH. The mixture was stirred at 40°C for 15 min to give a product that was washed with EtOH (yield 75%).

M.p. 4(5)methyl-2-mercaptoimidazoline decomposed at 129–160°C [32], 4(5)phenyl-2-mercaptoimidazoline at 191–193.5°C [33], and 4(5)benzyl-2-mercaptoimidazoline at 152–154°C [34].

4.1.6. General method of preparation of 4(5)-R-2-[ω-[4(5)imidazolyl]alkyl]thio]imidazolines (3a–3h)

The appropriate 2-mercaptoimidazoline (5 mmol) was added to a solution of 5.8 mmol of 4(5)-(ω-chloroalkyl)imidazole and 5.8 mmol of KI in the minimum amount of DMF. The mixture was stirred at 60°C for 20 h. The solvent was then evaporated under reduced pressure and the residue was purified by column chromatography (SiO_2, **3a**–**3d**: CH_2Cl_2–CH_3OH = 18:1; **3e**–**3h**: CH_2Cl_2–CH_3OH = 20:1). Yields and characteristic data of these compounds are reported in Table 1.

^{1}H NMR (DMSO-d_6) **3a** (dihydrochloride): δ 3.01 (t, 2H, CH_2–Im), 3.68 (t, 2H, CH_2–S), 3.85 (s, 4H, imidazoline), 7.57 (s, 1H, Im-5-H), 9.07 (s, 1H, Im-2-H).

^{1}H NMR (DMSO-d_6) **3b** (oxalate): δ 1.24 (d, 3H, CH_3), 2.93 (t, 2H, CH_2–Im), 3.37–3.48 (m, 3H, CH_2–S; imidazoline), 3.95 (app. t, J = 10.1 Hz, 1H, imidazoline), 4.23–4.38 (m, 1H, imidazoline), 7.06 (s, 1H, Im-5-H), 7.95 (s, 1H, Im-2-H).

^{1}H NMR (DMSO-d_6) **3c** (hydrochloride): δ 3.15 (t, 2H, CH_2–Im), 3.69 (m, 3H, CH_2–S; imidazoline), 4.37 (app. t, J = 11.1 Hz, 1H, imidazoline), 5.43 (dd, J = 11.1, J = 8.3 Hz, 1H, imidazoline), 7.42 (m, 5H, Ph), 7.59 (s, 1H, Im-5-H), 9.08 (s, 1H, Im-2-H).

^{1}H NMR (DMSO-d_6) **3d** (dihydrochloride): δ 2.94 (d, 2H, CH_2–Ph), 3.10 (t, 2H, CH_2–Im), 3.57–3.67 (m, 3H, CH_2–S; imidazoline), 3.88 (app. t, 1H, imidazoline), 4.54–4.64 (m, 1H, imidazoline), 7.31 (m, 5H, Ph), 7.54 (s, 1H, Im-5-H), 9.05 (s, 1H, Im-2-H).

^{1}H NMR (DMSO-d_6) **3e** (dihydrochloride): δ 2.05 (m, 2H, CH_2–CH_2–CH_2), 2.76 (t, 2H, CH_2–Im), 3.30 (t, 2H, CH_2–S), 3.84 (s, 4H, CH_2–imidazoline), 7.45 (s, 1H, Im-5-H), 8.99 (s, 1H, Im-2-H).

^{1}H NMR (DMSO-d_6) **3f** (oxalate): δ 1.19 (d, 3H, CH_3), 1.87 (m, 2H, CH_2–CH_2–CH_2), 2.94 (t, 2H, CH_2–Im), 3.35–3.45 (m, 3H, CH_2–S; imidazoline), 3.90–4.00 (m, 1H, imidazoline), 4.24–4.35 (m, 1H, imidazoline), 7.10 (s, 1H, Im-5-H), 8.06 (s, 1H, Im-2-H).

^{1}H NMR (DMSO-d_6) **3g** (free base): δ 1.93 (m, 2H, CH_2–CH_2–CH_2), 2.60 (t, 2H, CH_2–Im), 3.05 (t, 2H, CH_2–S), 3.27 (dd, J = 11.1, J = 8.2 Hz, 1H, imidazoline), 3.96 (app. t, J = 10.7 Hz, 1H, CH–imidazoline), 4.90 (app. t, J = 9.1 Hz, 1H, imidazoline), 6.76 (s, 1H, Im-5-H), 7.30 (m, 5H, Ph), 7.49 (s, 1H, Im-2-H).

^{1}H NMR (DMSO-d_6) **3h** (dioxalate): δ 1.64 (d, 2H, CH_2–Bz), 1.94 (m, 2H, CH_2–CH_2–CH_2), 2.70 (t, J = 7.1 Hz, 2H, CH_2–Im), 2.85–2.98 (m, 1H, imidazoline), 3.19 (t, J = 7.1 Hz, CH_2–S), 3.60 (dd, J = 11.0, J = 6.6 Hz, 1H, imidazoline), 3.87 (app. t, J = 10.8 Hz, 1H, imidazoline), 7.26 (s, 5H, Ph), 7.60 (s, 1H, Im-5-H), 8.43 (s, 1H, Im-2-H).

4.2. Pharmacology

4.2.1. Binding assays

Rat (Wistar) brain membranes were incubated for 30 min with [^{3}H]-RAMHA, in Tris–HCl 50 mM, pH 7.4, NaCl 50 mM, EDTA 0.5 mM and rapidly filtered under vacuum. Specific binding was defined as the binding inhibited by thioperamide 10 μM, and the pK_i values were calculated from the inhibition curves of the compounds tested versus 0.5 nM [^{3}H]-RAMHA according to Cheng and Prusoff's equation [36].

4.2.2. Functional assays

Portions of guinea-pig ileum were mounted on a coaxial platinum electrode assembly in a 10 ml water-jacketed organ-bath containing Krebs–Henseleit solution aerated with 95% O_2:5% CO_2 and maintained at 37°C. The preparation was then equilibrated for 60 min under 1 g of resting tension, with replacement of fresh solution every 15 min. Single electrical pulses were delivered to the tissue at 0.1 Hz frequency and 1 ms duration from a stimulator (LACE Elettronica model ES-3, Ospedaletto PI, Italy) with submaximal voltage (1.5–3.0 V). Cumulative concentration–response curves for the inhibition of electrically stimulated contractions were determined for the H_3 selective agonist RAMHA (1 nM–1 μM). The tissues were allowed to equilibrate with the compounds under study (1 nM–10 μM) for 30 min before the generation of concentration–response curves to the agonist. The antagonist potencies of the drugs tested were estimated using pA_2 determination as described by Arunlakshana and Schild [37]; otherwise, when only two high concentrations were tested, pK_B values ('apparent pA_2') were determined according to Furchgott's equation [38]:

$$pA_2 \text{ app} = \log([E']/[E] - 1) - \log[B]$$

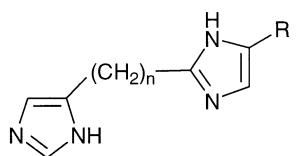
where [E'] and [E] are the concentrations of the agonist producing the half-maximum effect in the presence and

absence of the antagonist, respectively; [*B*] is the concentration of the antagonist.

5. Results and discussion

Biological data on H_3-receptor affinity and antagonist potency for newly synthesised 2-alkylimidazole derivatives, with alkyl spacers ranging from dimethylene to tetramethylene chains and phenyl or cyclohexyl lipophilic ending groups, are reported in Table 2, while those for ethylthio- and propylthio-imidazoline derivatives are reported in Table 3. Table 4 reports the biological data for thioimidazole derivatives [20,23,25,39] to be used for comparison.

Table 2
H_3-receptor affinity (pK_i) and antagonist activity (pA_2) of the 2-alkylimidazole derivatives



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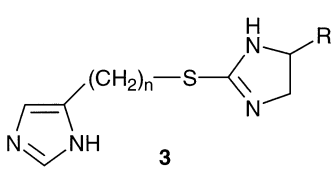
Comp.	*n*	R	pK_i	pA_2
2a	2	C_6H_5	7.74 ± 0.04	6.81 ± 0.11
2b	2	cC_6H_{11}	7.00 ± 0.07	8.09 ± 0.14
2c	3	C_6H_5	7.07 ± 0.20	8.48 ± 0.35
2d	3	cC_6H_{11}	7.90 ± 0.04	8.12 ± 0.30 [a]
2e	4	C_6H_5	8.03 ± 0.07	7.31 ± 0.09
2f	4	cC_6H_{11}	8.97 ± 0.06	8.45 ± 0.31

[a] pK_B value obtained according to Furchgott's method [38].

Table 3
H_3-receptor affinity (pK_i) and antagonist activity (pA_2) of the 2-alkylthioimidazoline derivatives



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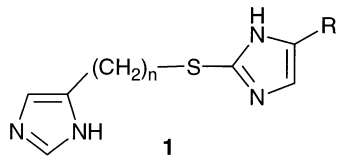
Comp.	*n*	R	pK_i	pA_2
3a	2	H	6.25 ± 0.10	6.78 ± 0.27
3b	2	CH_3	5.90 ± 0.08	6.68 ± 0.36
3c	2	C_6H_5	6.90 ± 0.11	[a]
3d	2	$CH_2C_6H_5$	7.63 ± 0.03	7.44 ± 0.17 [b]
3e [c]	3	H	6.53 ± 0.06	<6
3f	3	CH_3	7.32 ± 0.05	7.38 ± 0.35
3g	3	C_6H_5	6.98 ± 0.07	7.54 ± 0.35
3h	3	$CH_2C_6H_5$	7.53 ± 0.05	7.20 ± 0.30

[a] Non-competitive antagonism was observed at 10^{-7} M.
[b] pK_B value obtained according to Furchgott's method [38].
[c] α_2-Agonist, $pD_2 = 5.5$ (versus yohimbine).

Table 4
H_3-receptor affinity (pK_i) and antagonist activity (pA_2) of the 2-alkylthioimidazole derivatives



1

Comp.	*n*	R	pK_i	pA_2
1a	2	H	7.24 [23]	7.14 [39]
1b	2	CH_3	7.42 [20]	7.91 [20]
1c	2	C_6H_5	7.85 [25]	8.19 [25]
1d	2	cC_6H_{11}	$pK_{i,1} = 7.99$ [20] $pK_{i,2} = 9.89$ [20]	9.07 [20]
1e	2	$CH_2C_6H_5$	7.03 [20]	8.17 [20]
1f	3	H	6.52 [20]	<6 [20]
1g	3	CH_3	6.66 [20]	6.88 [20]
1h	3	C_6H_5	7.68 [20]	<6 [20]
1i	3	cC_6H_{11}	$pK_{i,1} = 6.76$ [20] $pK_{i,2} = 7.97$ [20]	7.27 [20]

The compounds tested behaved as H_3-antagonists on guinea-pig ileum, and displaced [^{3}H]-RAMHA from rat brain membranes with K_i values spanning three orders of magnitude, from millimolar to nanomolar values. Potency data on guinea-pig ileum (pA_2) generally confirmed the affinity values measured on rat brain membranes (pK_i), even if individual differences arose, probably due to the different biological procedures.

2-Alkylimidazole derivatives (**2a**–**f**), having the central heterocycle endowed with an intermediate basicity (its pK_a is expected to be close to 7), showed the best affinity values (pK_i 7.00–8.97), particularly with the longest tetramethylene chain. In contrast, the basic imidazoline derivatives (**3a**–**h**) gave poorer affinity values (pK_i 5.90–7.63), with a complex SAR pattern.

For the latter compounds (**3a**–**h**), the interpretation of the biological data is not straightforward, nor can a clear preference between the ethylthio and the propylthio chains be stated, although the positive influence of the 4(5)-lipophilic group, which had been observed for 2-thioimidazole derivatives **1a**–**i**, can still be recognised. Starting from very similar pK_i values for the parent compounds, **3a** and **3e**, the phenyl group gave a slight improvement in affinity, although this was not sufficient to reach good pK_i values. As this group steps out of the imidazoline ring plane, we tried to reduce the out-of-plane hindrance through the synthesis of a 4(5)-methyl and of a flexible 4(5)-benzyl derivative. Slight, but significant, improvement in affinity was obtained on the whole, although the best pK_i values, achieved for the benzyl derivatives, hardly reached those obtained for 2-alkylthioimidazole derivatives (**1a**–**i**).

Significantly higher affinities were obtained for 2-alkylimidazoles (**2a**–**f**). In this series the cyclohexyl

group, introduced in order to gently increase the imidazole basicity with respect to the phenyl derivative, led to a significant improvement of affinity, at least in the presence of the three- and four-methylene chains. In particular, the longest spacer led to a compound (**2f**) endowed with nanomolar affinity ($K_i = 1.07$ nM).

While in this series the best results were obtained for the butyl spacer, topologically corresponding to the propylthio one in the series **1** and **3**, an opposite trend was observed for the neutral 2-alkylthioimidazole derivatives (**1a**–**i**), where the ethylthio chain gave higher affinities than the propylthio one, irrespective of the lipophilic substituent.

Polar group basicity therefore seems to influence the optimal length of the chain spacer: longer chains are preferred in the presence of basic polar groups, as it has been observed for 2-alkylimidazoles (**2a**–**f**) and for the clobenpropit-like isothiourea derivatives [26], whereas shorter ones give higher affinities for the neutral 2-alkylthioimidazole derivatives **1a**–**i**.

While a moderate increase in the imidazole polar group basicity is well tolerated, the basic imidazoline derivatives (**3a**–**h**) gave the lowest affinity values among the three series, and were not able to reproduce the high H_3-potencies observed for the similar isothiourea derivatives [26], probably owing to an unfavourable orientation of the lipophilic group, forced by the ring closure.

These observations indicate that polar groups of intermediate basicity, such as the 2-alkylimidazole one, can confer high affinity for the H_3-receptor; they should be preferred to strongly basic ones, because a significant unionised fraction at physiological pH may be necessary to cross lipophilic barriers. Such groups should therefore be taken into consideration for the design of new potent H_3-antagonists active at the CNS.

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