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Non-imidazole histamine H_3 ligands. Part I. Synthesis of 2-(1-piperazinyl)- and 2-(hexahydro-1*H*-1,4-diazepin-1-yl)benzothiazole derivatives as H_3-antagonists with H_1 blocking activities

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Abstract

New 2-(1-Piperazinyl)- and 2-(hexahydro-1*H*-1,4-diazepin-1-yl)benzothiazoles were prepared and tested as H_1- and H_3-receptor antagonists. A number of compounds showed weak H_1-antagonistic activity, with pA_2 values ranging from 5.5 to 6.1. The simple alkyl substituted, 2-[1-(4-methyl and 4-ethyl)piperazinyl] analogues show increasing, moderate H_3-antagonistic activity ($pA_2 = 6.0$, and $pA_2 = 7.0$). The compounds with 4-phenylalkyl substitution, for both the piperazinyl and the hexahydro-1*H*-1,4-diazepin-1-yl homologues series, regardless of the different physicochemical properties of the *para* substituents at the phenyl ring, showed weak H_3-antagonistic activity with pA_2 values ranging from 4.4 to 5.6. © 1999 Elsevier Science S.A. All rights reserved.

Keywords: Histamine H_1- and H_3-receptors; H_1- and H_3-antagonists; 2-Piperazinyl- and 2-(hexahydro-1*H*-1,4-diazepin-1-yl)benzothiazoles

1. Introduction

Histamine plays a key role in allergy and inflammation (via H_1-receptors) and in gastric secretion (via H_2-receptors) [1]. The H_1-receptor has been a target for drug discovery for many years, and H_1-receptor antagonists have proved to be effective therapeutic agents for the treatment of allergic rhinitis. However, classical antihistamine agents have several limitations which complicate their clinical use, such as non-selective pharmacological activity and central nervous system (CNS) activity. H_1-Antagonists (promethazine, diphenhydramine, cyclazine) demonstrate muscarine-receptor antagonist activity, which may produce anticholinergic side effects. The sedative activity of H_1-antagonists is associated with binding to cerebral H_1-receptors [2]. The focus of newer H_1-antagonists has been an efficacy with diminished sedative liability. These agents are used in rhinitis, hay fever, asthma, and obstructive airway disease [3–6]. As opposed to classical antihistamine, the more recent H_1-antagonists loratadine [7], astemizole [8], and temelastine [9], have poor access to the central nervous system (CNS) which produces non-sedating antihistaminic activity in the clinic. However, since the late 1980's, reports [10] began to appear indicating that patients who took intentional overdoses of terfenadine or astemizole could develop a classical form of ventricular arrhythmia, *torsades de pointes*, which has been previously associated with quinidine and other antiarrhythmic drugs. Many H_1-antihistamines have now been examined for their cardiac actions. Astemizole and terfenadine, among others, belong to a group of antihistamines with cardiac effects at their antihistamine concentrations [11–13]. New, non-sedative H_1-antagonists are therefore still needed.

More recently, in addition to these two postsynaptic receptor subtypes, presynaptic H_3-receptors have been identified [14] in the brain. These receptors were described to be located presynaptically on histaminergic

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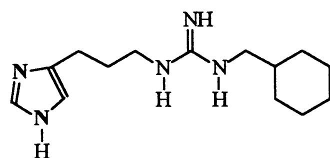
nerve endings, regulating the release and synthesis of histamine by a negative feedback (autoreceptor). The histamine H_3-receptor has since been shown, not only to inhibit the synthesis and release of histamine, but also to play an important regulatory role in the release of other neurotransmitters (e.g. serotonin, acetylcholine, noradrenaline) in the CNS [15–18] and in the periphery as well (heteroreceptors) [19–23]. Moreover, H_3-receptors are also present in a variety of peripheral sites such as the cardiovascular, respiratory and gastrointestinal systems [24–28]. Confirmation of the existence of the third histamine receptor subtype was provided by the development of the H_3-selective agonist (R)-α-methylhistamine and the H_3-selective antagonist thioperamide [29].

In recent years the histamine H_3-receptor has become of great interest. It has been postulated after extensive in vitro and in vivo research that the H_3-receptor might be involved in several neuronal diseases like Alzheimers, epilepsy or schizophrenia [30–33].

It has been shown that potent H_3-receptor antagonists, known so far, are all imidazole derivatives connected with a polar group by a chain. This polar group is connected with lipophilic moiety [34], by another chain.

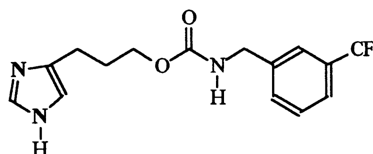
In most classes of histamine H_3-receptor antagonists, three methylene groups were found to be the optimum for chain connecting 4-imidazolyl moiety with a polar group. The diversity of the polar group (i.e. amines, guanidines, amides, thioamides) leads to the conclusion that the basicity is of no importance in the activity of these compounds. There are many possibilities for variation of the side chain. The reported H_3-antagonists may be classified into the following groups: amine derivatives **1** [35], carbamates **2** [36], ethers **3** [37], heteroaryl derivatives **5**, **6** [38,39] and less polar alkenes

Amine Derivatives



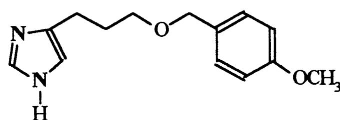
1; $-\log K_i = 9.1^a$

Carbamates



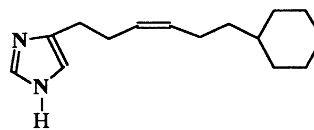
2; $-\log K_i = 8.1^b$

Ethers



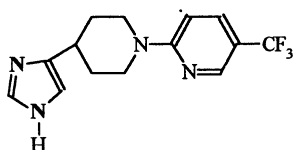
3; $-\log K_i = 8.3^b$

Alkene Derivatives

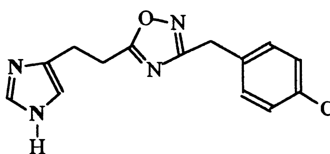


4; $-\log K_i = 8.4^c$

Heteroaryl Derivatives

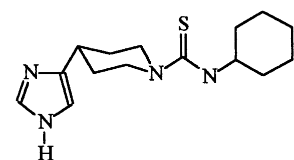


5; $-\log K_i = 7.4^c$

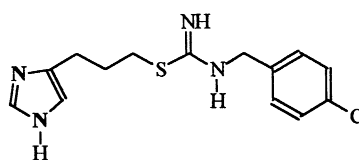


6; $-\log K_i = 8.2^b$

Thioureas and Isothioureas



7; $-\log K_i = 8.3^d$



8; $pA_2 = 9.9^e$

Scheme 1. The imidazole derivatives as potent H_3-receptor antagonists. Functional assay on synaptosomes of rat cerebral cortex: a [29], b [65], c [40], d [14]; functional assay on guinea pig ileum: e [20].

4 [40] (Scheme 1) and also alkynes derived from the marine natural product verongamine [41].

A number of potent and selective H_3-receptor antagonists possess sulfur-containing functionalities, e.g. the thiourea derivatives thioperamide **7** [20], widely used in experimental studies, and the highly potent isothiourea derivative clobenpropit **8** [42] (Scheme 1).

There are only a few non-imidazoles such as betahistine [43], phencyclidine [44], dimaprit [45,46], and clozapine [47,48] having rather weak, but pertinent antagonist activity.

Very recently, Ganellin et al. [49] reported a novel series of homologues *O* and *S* isosteric tertiary amines of *N*-ethyl-*N*-(4-phenylbutyl)amine as potent non-imidazole histamine H_3-receptor antagonists.

Our starting point, based on the previous literature study, was the observation that the benzothiazolyl–imidazolyl–piperidine derivatives [50] showed moderate to good histamine H_3-receptor antagonist activity.

In order to further investigate the structural requirements for the H_3-receptor and to search for possible non-imidazole ligands, two series of benzothiazole (as a pseudo isothiourea group) derivatives, where the imidazole ring in 2-benzothiazolyl-4-[(1*H*)imidazol-4-yl] derivatives was replaced with other heterocyclic rings (piperazine or homopiperazine), were synthesized and evaluated for their antagonistic histamine H_3-activity.

The aim of the present work was the synthesis and pharmacological in vitro evaluation of histamine H_3-receptor antagonists being free of any imidazole-containing functionality. The new 2-(1-piperazinyl)benzothiazole and 2-(hexahydro-1*H*-1,4-diazepin-1-yl)benzothiazole derivatives (Tables 1 and 2) are endowed both with H_1- (as a pseudo analogue of 2-(4-substituted-1-piperazinyl)- and 2-(4-substituted-1-homopiperazinyl)-benzimidazoles, potent H_1-receptor antagonist in vitro and in vivo) [51] and H_3-antagonist properties.

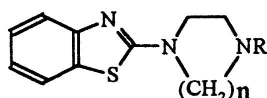
2. Results and discussion

The presented simple alkyl-substituted piperazine analogues **10a**,**b** show increasing, moderately potent H_3-receptor antagonistic activity. Replacement of the piperazine ring by hexahydro-1*H*-1,4-diazepine leads to the compounds **11a** and **11b** with weaker biological activity. The benzyl and phenethyl derivatives of 2-piperazinylbenzothiazole (**10c**–**j**), independently on substituent in *para* position of the phenyl ring, possess weak activity. The corresponding 2-(hexahydro-1*H*-1,4-diazepin-1-yl)benzothiazole (**11c**–**f**,**i**), analogues of piperazine series, are compounds with poor H_3-receptor antagonist activity; in the case of compounds **17g**,**h**,**j** the activity is completely lost.

Compound **10b** ($pA_2 = 7.02$) is the most effective H_3-receptor antagonist of this series. Further investigations are necessary to clarify the dependency, within the homologue series of simple alkyl substituents of 2-piperazinylbenzothiazoles, of activity on the size of the piperazinyl substituent.

A number of the series of compounds showed weak, but competitive H_1-antagonistic activity, with pA_2 values ranging from 5.5 to 6.1.

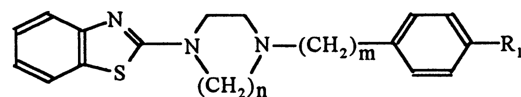
Table 1
Reaction details and physical data for compounds **10**, **10a**,**b** and **11**, **11a**,**b** and corresponding dihydrobromides



Comp.	R	*n*	M.p. (°C)	TLC, R_f, and index of eluting mixture [c]	Reaction		Yield (%)	Dihydrobromides	
					Time (h)	Temperature (°C)		Formula [d]	M.p. (°C)
10 [a]	H	2	75–77 [52]	0.41, a	12	reflux	92	$C_{11}H_{15}Br_2N_3S$	288–289
10a [a]	CH_3	2	94–95 [52]	0.41, h	24	reflux	52	$C_{12}H_{17}Br_2N_3S$	276–278
10b [b]	CH_2CH_3	2	108–110	0.33, g	24	66	58	$C_{13}H_{19}Br_2N_3S$	274.5–275.5
11 [a]	H	3	73–75 [59]	0.31, b	12	reflux	85	$C_{12}H_{17}Br_2N_3S$	267–269
11a [a]	CH_3	3	sticky oil	0.53, f	24	reflux	45	$C_{13}H_{19}Br_2N_3S$	262–264
11b [b]	CH_2CH_3	3	sticky oil	0.21, f	18	66	62	$C_{14}H_{21}Br_2N_3S$	287–288

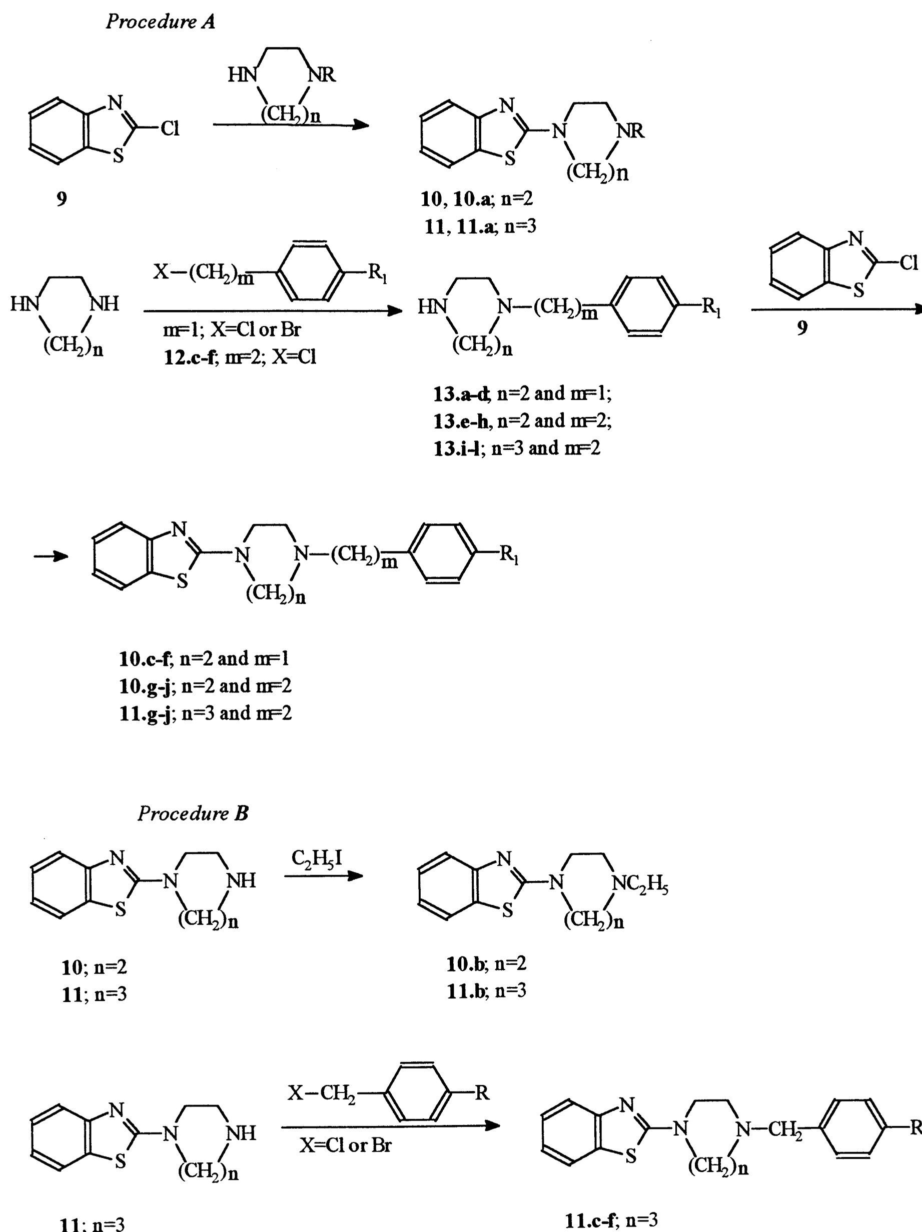
[a] Reaction solvent: 80% 2-PrOH with presence of $NaHCO_3$.
[b] Reaction solvent: benzene.
[c] See Section 4.
[d] Analytical results for C, H, N were within $\pm 0.3\%$ of the calculated values.

Table 2
Reaction details and physical data for compounds **10c–j**, **11c–j** and corresponding dihydrobromides



Comp.	R_1	*n*	*m*	M.p. (°C)	TLC, R_f, and index of eluting mixture [c]	Crystallization solvent	Reaction		Yield (%)	Dihydrobromides	
							Time (h)	Temp. (°C)		Formula [d]	M.p. (°C)
10c [a]	H	2	1	136.5–137.5	0.19, e	acetone	18	reflux	90.6	$C_{18}H_{21}Br_2N_3S$	250–252
10d [a]	CH_3	2	1	171.5–172.5	0.29, e	acetone	24	reflux	69	$C_{19}H_{23}Br_2N_3S$	260–262
10e [a]	OCH_3	2	1	165.5–166.5	0.25, e	acetone	24	reflux	70.8	$C_{19}H_{23}Br_2N_3OS$	245–246
10f [a]	$C(CH_3)_3$	2	1	134.5–136	0.35, e	acetone	24	reflux	47	$C_{22}H_{29}Br_2N_3S$	261–263
10g [a]	H	2	2	142–143.5	0.20, e	*n*-propanol	36	reflux	55.5	$C_{19}H_{23}Br_2N_3S$	267–269
10h [a]	CH_3	2	2	125.5–127	0.26, e	*n*-propanol	36	reflux	69	$C_{20}H_{25}Br_2N_3S$	277–279
10i [a]	OCH_3	2	2	130–131.5	0.23, e	*n*-propanol	36	reflux	65	$C_{20}H_{25}Br_2N_3OS$	255–257
10j [a]	$C(CH_3)_3$	2	2	132–133	0.28, e	*n*-propanol	36	reflux	76	$C_{23}H_{31}Br_2N_3S$	302–304
11c [b]	H	3	1	107–108	0.80, b		1	75	31	$C_{19}H_{23}Br_2N_3S$	223–225
11d [b]	CH_3	3	1	82–83	0.80, b		1	80	57	$C_{20}H_{25}Br_2N_3S$	234–235
11e [d]	OCH_3	3	1	80–81	0.77, b		1	80	26	$C_{20}H_{25}Br_2N_3OS$	189.5–190
11f [b]	$C(CH_3)_3$	3	1	102–103	0.82, b		1	85	58	$C_{23}H_{31}Br_2N_3S$	222–223
11g [a]	H	3	2	63–64	0.78, b		24	reflux	50	$C_{20}H_{25}Br_2N_3S$	246–248
11h [a]	CH_3	3	2	75–77	0.77; b		24	reflux	43	$C_{21}H_{27}Br_2N_3S$	255–256
11i [a]	OCH_3	3	2	87–88	0.75, b		24	reflux	41	$C_{21}H_{27}Br_2N_3OS$	253–254
11j [a]	$C(CH_3)_3$	3	2	sticky oil	0.80, b		24	reflux	84	$C_{24}H_{33}Br_2N_3S$	250–251

[a] Reaction solvent: 80% 2-PrOH with presence of $NaHCO_3$.
[b] Reaction solvent: *N*,*N*-dimethylformamide in the presence of excess $N(C_2H_5)_3$.
[c] See Section 4.
[d] Analytical results for C, H, N were within $\pm 0.3\%$ of the calculated values.



Scheme 2.

3. Chemistry

The general synthetic procedures used in this study are illustrated in Scheme 1.

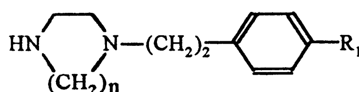
The 2-(1-piperazinyl- and 2-(1-homopiperazinyl[1])-benzothiazole derivatives (**10**, **10a**, **10c**–**j**, **11** [59], **11a** and **11g**–**j**) were obtained, according to the procedure of Verderame [52], from 2-chlorobenzothiazole (**9**) through nucleophilic substitution of the chlorine atom by the appropriate piperazine or homopiperazine in the presence of sodium bicarbonate in 80% 2-propanol (Scheme 2, procedure A).

[1] In chemical section, instead of hexahydro-1*H*-1,4-diazepine, the colloquial name of this compound is used — homopiperazine.

Derivatives **10b** and **11b**, (Scheme 2, procedure B) were synthesized from 2-(1-piperazinyl)benzothiazole (**10**) and 2-(1-homopiperazinyl)benzothiazole (**11**) by alkylation with the ethyl iodide; derivatives **11c**–**f** (Scheme 2, procedure B) were obtained from 2-(1-homopiperazinyl)benzothiazole (**11**) by reaction with the corresponding 4-*R*-benzyl halide.

The following monosubstituted piperazines **13a**–**e**,**g** (Scheme 2, procedure A) were prepared by literature methods: 1-(4-benzyl)piperazine [60], 1-(4-(4-methyl)-benzyl)piperazine [60], 1-(4-(4-methoxy)benzyl)piperazine [60], 1-[4-(4-(*tert*-butyl))benzyl]piperazine [61], 1-(4-phenethyl)piperazine [62], 1-(4-(4-methoxy)-phenethyl)piperazine [63]. The monosubstituted piperazines **13f**,**h** and monosubstituted homopiperazines

Table 3
Reaction details and physical data for compounds **13f**,**h**–**l**



Comp.	R_1	*n*	Formula mol.wt. (g/mol)	TLC, R_f of free base, and index of eluting mixture	B.p./mm Hg (°C)	Reaction time (h)	Yield (%)
13f [a]	CH_3	2	$C_{13}H_{20}N_2$	0.40, d		48	49
13h [a]	$C(CH_3)_3$	2	$C_{16}H_{26}N_2$	0.41, d		48	51
13i [b]	H	3	$C_{13}H_{20}N_2$	0.12, d	136–138/10	24	46
13j [b]	CH_3	3	$C_{14}H_{22}N_2$	0.15, d	178–179/10	24	56
13k [b]	OCH_3	3	$C_{14}H_{22}N_2O$	0.09, d	132–134/0.2	24	49
13l [b]	$C(CH_3)_3$	3	$C_{17}H_{28}N_2$	0.19, d		24	51

[a] Reaction solvent: toluene.
[b] Reaction solvent: ethanol.

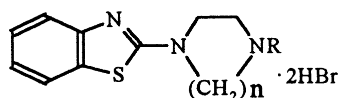
13i–**l** (Scheme 1, procedure A) were obtained by the reaction of five equivalents piperazine or homopiperazine with one equivalent of the appropriate benzyl chloride or bromide or phenethyl chloride in toluene or ethyl alcohol (Table 3).

The results concerning compounds of Tables 1 and 2 are collected, respectively, in Tables 4 and 5.

The 2-chlorobenzothiazole, benzyl bromide, 4-methyl, 4-methoxy-, 4-(*tert*-butyl)benzyl chlorides, (2-chloroethyl)benzene, 4-methyl-, 4-methoxyphenethyl alcohols, 1-methylpiperazine, 1-methylhomopiperazine, and ethyl iodide were purchased from commercial sources. The 4-methyl- [53], 4-methoxy- [54] and 4-(*tert*-butyl)-phenethyl [55] chlorides (**12a**–**c**) (Scheme 2) were directly obtained by the reaction of the corresponding alcohol with an excess of thionyl chloride.

The 4-(*tert*-butyl)phenethyl alcohol (**16c**) [56] (Scheme 3) was obtained via the following steps: methyl 4-(*tert*-butyl)phenyl acetate (**15**) [57,58] was obtained by reaction of sodium cyanide on 4-(*tert*-butyl)benzyl chloride and treatment of obtained 4-(*tert*-butyl)benzyl cyanide (**14**) [57] with methanol in the presence of *p*-toluenesulfonic acid monohydrate. In the last step the ester was converted into 4-(*tert*-butyl)phenethyl alcohol (**16c**) by reduction of **15** with lithiumaluminum hydride in dry ethyl ether (Scheme 2).

Table 4
H_1- and H_3-antagonistic activity of compounds **10**, **10a**,**b** and **11**, **11a**,**b** as tested on the in vitro test system on the guinea pig jejunum



Comp.	R	*n*	pA_2 (SEM)		pA_2 (SEM)	
			H_1 [a]	*N* [b]	H_3	*N* [b]
10	H	2	5.82 (0.04)	8	5.43 (0.07)	5
10a	CH_3	2	5.60 (0.06)	8	5.95 (0.06)	5
10b	CH_2CH_3	2	5.77 (0.04)	6	7.02 (0.03)	5
11	H	3	5.50 (0.05)	7	5.11 (0.21)	5
11a	CH_3	3	5.70 (0.04)	4	5.96 (0.20)	5
11b	CH_2CH_3	3	NT		6.79 (0.21)	5

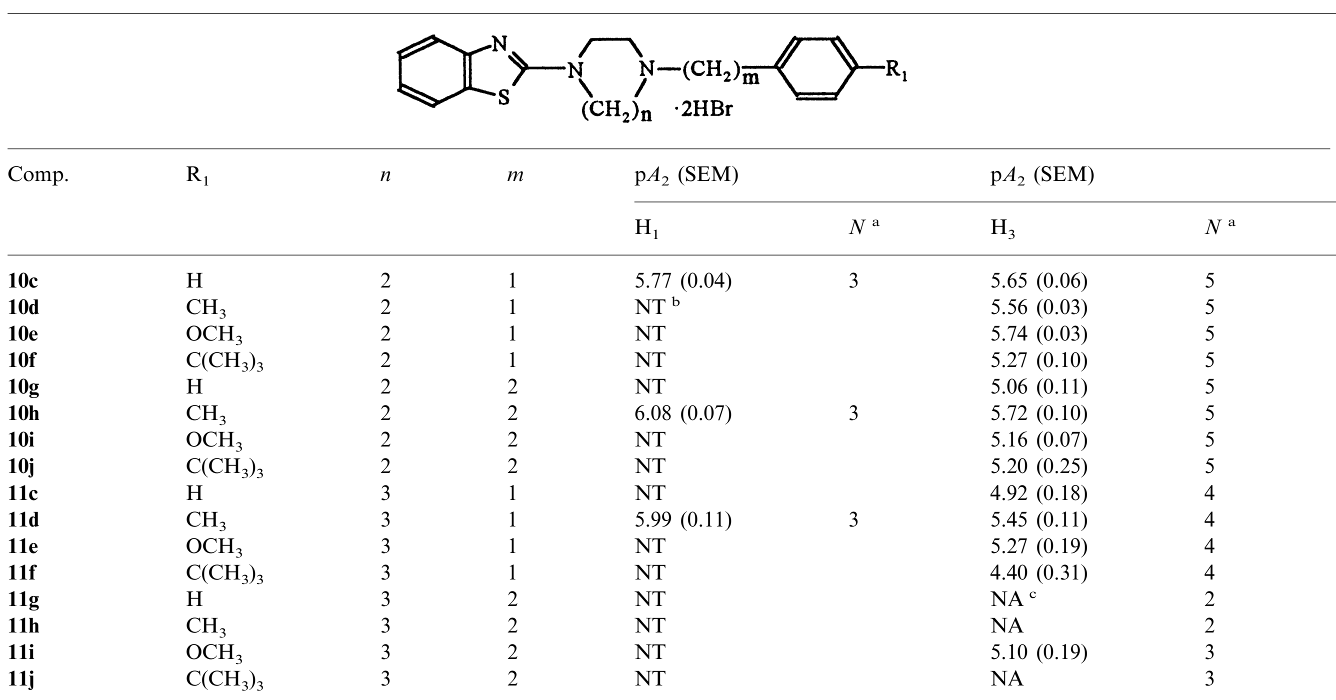
[a] NT, not tested.
[b] Number of different animal preparations.

4. Experimental

4.1. General methods

All melting points (m.p.) were taken in open capillaries on an electrothermal apparatus and are uncorrected. For all compounds ^1H NMR spectra were recorded on a Varian EM 360 (60 MHz) spectrometer. Chemical shifts are expressed in ppm downfield from internal TMS as reference. ^1H NMR data are reported in the following order: multiplicity (br, broad; s, singlet; d, doublet; t, triplet; m, multiplet; *, exchangeable by D_2O) number of protons, and approximate coupling constant in Hertz. Elemental analyses (C, H, N) for all compounds were performed on Heraeus EA 415-0 and the results are within $\pm$ 0.3% of the theoretical values. TLC was performed on Silica Gel PF_{254} plates (E. Merck), using the following eluting mixtures: (a) 89.8:10:0.2 methylene chloride–methanol–concentrated ammonium hydroxide; (b) 90:10:1 methylene chloride–methanol–concentrated ammonium hydroxide; (c) 89:10:1 methylene chloride–methanol–concentrated ammonium hydroxide; (d) 88:20:2 methylene chloride–methanol–concentrated ammonium hydroxide; (e) 39:1 methylene chloride–methanol; (f) 20:1 methylene chloride–methanol; (g) 14:1 methylene chloride–methanol; and (h) 9:1 methylene chloride–methanol. Column chromatography was carried out using Silica Gel 30–60 μm (J.T. Baker), employing the same eluent as was indicated by TLC.

Table 5
H_1- and H_3-antagonistic activity of compounds **10c–j** and **11c–j** as tested on the in vitro test system on the guinea pig jejunum



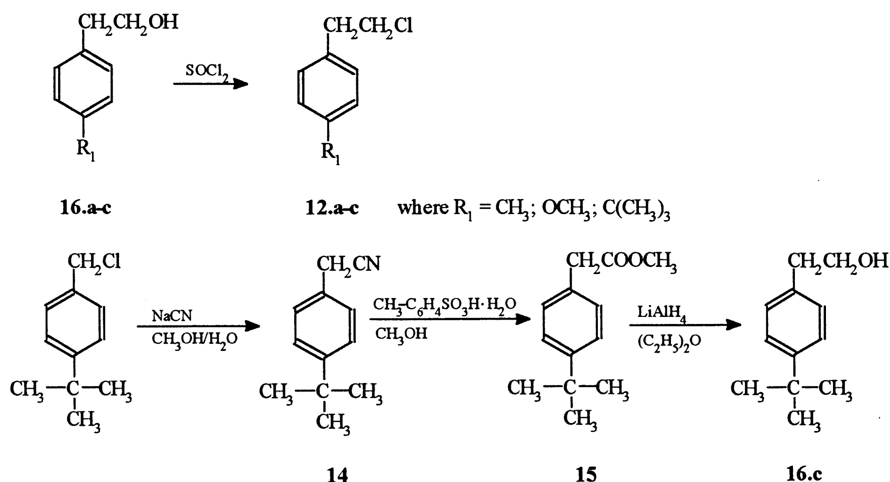
Comp.	R_1	*n*	*m*	pA_2 (SEM)		pA_2 (SEM)	
				H_1	*N* [a]	H_3	*N* [a]
10c	H	2	1	5.77 (0.04)	3	5.65 (0.06)	5
10d	CH_3	2	1	NT [b]		5.56 (0.03)	5
10e	OCH_3	2	1	NT		5.74 (0.03)	5
10f	$C(CH_3)_3$	2	1	NT		5.27 (0.10)	5
10g	H	2	2	NT		5.06 (0.11)	5
10h	CH_3	2	2	6.08 (0.07)	3	5.72 (0.10)	5
10i	OCH_3	2	2	NT		5.16 (0.07)	5
10j	$C(CH_3)_3$	2	2	NT		5.20 (0.25)	5
11c	H	3	1	NT		4.92 (0.18)	4
11d	CH_3	3	1	5.99 (0.11)	3	5.45 (0.11)	4
11e	OCH_3	3	1	NT		5.27 (0.19)	4
11f	$C(CH_3)_3$	3	1	NT		4.40 (0.31)	4
11g	H	3	2	NT		NA [c]	2
11h	CH_3	3	2	NT		NA	2
11i	OCH_3	3	2	NT		5.10 (0.19)	3
11j	$C(CH_3)_3$	3	2	NT		NA	3

[a] Number of different animal preparations.
[b] NT, not tested.
[c] NA, inactive.

*4.2. General method for the preparation of 2-(1-piperazinyl), and 2-(1-homopiperazinyl)benzothiazoles (**10**, **10a**, **10c–j**, **11**, **11a** and **11g–j**)*

To a refluxing mixture of the corresponding piperazine or homopiperazine (0.01 mol) and sodium bicarbonate (0.02 mol) in 70 ml of 80% 2-PrOH was added dropwise a solution containing 2-chlorobenzothiazole (**9**) (0.005 mol) in 4 ml of 2-PrOH. The mixture was refluxed for 12–48 h. The solvent was evaporated under reduced pressure, and the semisolid residue was suspended in 40 ml of water. After stirring for 1 h, the mixture was extracted with CH_2Cl_2, and the solvent was removed in vacuo. The products were purified by column chromatography or recrystallized twice from acetone or *n*-propanol (Tables 1 and 2).

10a: $C_{12}H_{15}N_3S$ (233); ^1H NMR ($CDCl_3$ + TMS): δ 2.25 (s, 3H, CH_3), 2.45–2.60 (t, $4H_{piperazine}$), 3.55–3.80 (t, $4H_{piperazine}$), 7.0–7.6 (m, 4H, arom).



Scheme 3.

10d: $C_{19}H_{21}N_3S$ (323); ^{1}H NMR ($CDCl_3$ + TMS): δ 2.35 (s, 3H, CH_3), 2.50–2.65 (t, $4H_{piperazine}$), 3.55 (s, 2H, CH_2Ph), 3.60–3.75 (t, $4H_{piperazine}$), 7.0–7.6 (m, 8H, arom).

10i: $C_{20}H_{23}N_3OS$ (353); ^{1}H NMR ($CDCl_3$ + TMS): δ 2.50–2.90 (m, 8H, $4H_{piperazine}$, 4H, CH_2CH_2), 3.55–3.70 (t, $4H_{piperazine}$), 3.75 (s, 3H, OCH_3), 6.75–7.00 (m, 2H, arom), 7.1–7.50 (m, 4H, arom), 7.55–7.75 (m, 2H, arom).

11a: $C_{12}H_{15}N_3S$ (233); ^{1}H NMR ($CDCl_3$ + TMS): δ 1.95–2.15 (m, 2H, $CCH_2C_{homopiperazine}$), 2.45 (s, 3H, CH_3), 2.85–3.10 (m, $4H_{homopiperazine}$), 3.75–3.95 (m, $4H_{homopiperazine}$), 7.15–7.65 (m, 4H, arom).

11i: $C_{21}H_{25}N_3OS$ (367); ^{1}H NMR ($CDCl_3$ + TMS): δ 1.95–2.10 (m, 2H, $CCH_2C_{homopiperazine}$), 2.50 (s, 3H, OCH_3), 2.70–3.00 (m, 8H, $4H_{homopiperazine}$, 4H, CH_2CH_2Ph), 3.75–4.05 (m, $4H_{homopiperazine}$), 7.00–7.30 (m, 7H, arom), 7.50–7.70 (m, 2H, arom).

4.3. General method for the preparation of 2-[1-(4-ethyl)piperazinyl]-, and 2-[1-(4-ethylhomopiperazinyl]-benzothiazoles and 2-[1-(4-benzyl)homopiperazinyl]-benzothiazoles (*10b*, *11b* and *11c–f*)

To a solution of the corresponding 2-(1-piperazinyl- or 2-(1-homopiperazinyl)benzothiazole (**10** or **11**) (0.01 mol) dissolved in 250 ml of an appropriate solvent was added ethyl iodide (0.005 mol) or the corresponding *p*-benzyl halide (0.004 mol). The reaction mixture was heated for an appropriate time and at an appropriate temperature (Tables 1 and 3). After cooling, the solvent was evaporated and the residue was suspended in 50 ml of water. After stirring for 20 min, the mixture was extracted with CH_2Cl_2, the organic layer was separated, dried, and evaporated to give the crude product which was purified by column chromatography (Tables 1 and 2).

10b: $C_{13}H_{17}N_3S$ (247); ^{1}H NMR ($CDCl_3$ + TMS): δ 1.00–1.20 (t, $J = 7$ Hz, 3H, CH_3); 2.35–2.60 (m, 6H, $4H_{piperrazine}$, 2H, CH_2), 3.60–3.80 (t, $4H_{piperrazine}$); 7.0–7.6 (m, 4H, arom).

11b: $C_{14}H_{19}N_3S$ (261); ^{1}H NMR ($CDCl_3$ + TMS): δ 1.05–1.25 (t, $J = 7$ Hz, 3H, CH_3) 1.90–2.20 (m, 2H, $CCH_2C_{homopiperazine}$), 2.60–2.80 (m, 6H, $4H_{homopiperazine}$, 2H, CH_3 CH_2), 3.75–3.95 (m, $4H_{homopiperazine}$), 7.15–7.65 (m, 4H, arom).

11f: $C_{23}H_{29}N_3S$ (379); ^{1}H NMR ($CDCl_3$ + TMS): δ 1.30 (s, 9H, $3CH_3$), 1.90–2.15 (m, 2H, $CCH_2C_{homopiperazine}$), 2.70–2.95 (m, 8H, $4H_{homopiperazine}$, 2H, CH_2Ph), 3.70–3.90 (m, $4H_{homopiperazine}$), 7.05–7.40 (m, 6H, arom), 7.50–7.70 (m, 2H, arom).

All free bases were treated with methanolic HBr and hydrobromides were precipitated with dry diethyl ether.

4.4. General method for the preparation of 1-(4-benzyl)-, 1-(4-phenethyl)piperazines and 1-(4-phenethyl)homopiperazines (*13a–h* and *13i–l*)

To a solution of piperazine (in toluene, 400 ml) or homopiperazine (in ethyl alcohol, 150 ml) (0.2 mol) was added the appropriate substituted benzyl or phenethyl chloride (0.004 mol). The reaction mixture was refluxed for 24–48 h. The solvent was evaporated and 250 ml of a 10% solution of hydrochloric acid was added to the residue and the resulting mixture was extracted with diethyl ether. The organic layer was discarded and the water solution was made alkaline (pH ~ 14) and extracted with diethyl ether. The organic layer was separated, dried, and evaporated to give the crude product which was distilled under reduced pressure or purified by column chromatography (Table 3).

13h: $C_{16}H_{26}N_2$ (246); ^{1}H NMR ($CDCl_3$ + TMS): δ 1.30 (s, 9H, $C(CH_3)_3$), 2.15 (s*, 1H, NH), 2.45–2.70 (m, 8H, $4H_{piperrazine}$, 4H, CH_2CH_2Ph), 2.75–3.00 (t, $4H_{piperrazine}$), 7.25–7.50 (m, 4H, arom).

13l: $C_{17}H_{28}N_2$ (260); ^{1}H NMR ($CDCl_3$ + TMS): δ 1.30 (s, 9H, $(CH_3)_3$), 1.75–1.80 (m, 2H, $CCH_2C_{homopiperrazine}$), 1.85 (s*, 1H, NH); 1.90–2.10 (m, 2H, $CCH_2C_{homopiperrazine}$), 2.85–3.05 (m, 12 H, $8H_{homopiperrazine}$, 4H, CH_2CH_2Ph); 7.05–7.30 (m, 4H, arom).

4.5. 4-(tert-Butyl)phenethyl alcohol (*16c*)

To a refluxing mixture of (0.28 mol) pulverized lithium aluminum hydride in 150.0 ml of dry ether was added a solution of methyl 4-(*tert*-butyl)-phenylacetate (**15**) (0.46 mol) in 100.0 ml of dry ether at such a rate that the solvent refluxed gently. When the addition was complete, the mixture was stirred at the reflux temperature for an additional 30 min. The excess of lithium aluminum hydride was decomposed by adding water slowly with stirring. The precipitated inorganic salt was filtered off, and the solution was made acidic by addition of 10% aqueous hydrochloric acid. The organic layer was dried over sodium sulfate and evaporated. The residue was distilled under vacuum.

16c: $C_{12}H_{18}O$ [51] (178); b.p. 138–14°C/10 mmHg. Yield: 87%. ^{1}H NMR: ($CDCl_3$ + TMS): δ 1.30 (s, 9H, $C(CH_3)_3$), 2.05 (s*, 1H, OH), 2.70–2.85 (t, $J = 8$ Hz, 2H, $PhCH_2$), 3.75–2.9 (t, $J = 8$ Hz, 2H, CH_2OH), 7.15–7.40 (m, 4H, arom).

5. Pharmacology

All compounds were tested for H_1-antagonistic effects in vitro, following standard methods, using the guinea pig ileum [64].

Male guinea pigs weighing 300–400 g were sacrificed by a blow to the head. The ileum was excised and placed in phosphate buffer at room temperature (pH 7.4) containing (mM) NaCl (136.9), KCl (2.68), and $NaHPO_4$ (7.19). After flushing the intraluminal contents, segments of about 2 cm long were cut and mounted for isotonic contractions in water-jacked 20 ml organ baths filled with oxygenated (95:5 O_2–CO_2) Krebs buffer containing (mM) NaCl (117.5), KCl (5.6), $MgSO_4$ (1.18), $CaCl_2$ (2.5), NaH_2PO_4 (1.28), $NaHCO_3$ (25), and glucose (5.5) at 37°C under a constant load of 0.5 g. After a 30 min equilibration period with washings every 10 min, a sub maximal priming dose of histamine (1 μM) was given and washed out (standard washing procedure: three changes of buffer during 30 min). After washing out, the antagonistic activity of the given compounds was measured by recording a concentration–response curve (CRC) for histamine in the presence of the testing compounds (**10**, **10a**, **10b**, **10c**, **10g**, **11**, **11a** and **11e**) which were added 5 min before histamine. This procedure was repeated with higher concentrations of the compounds. The antagonism was of a competitive nature causing a parallel shift of the CRC. The pA_2-values were calculated according to Arunlakshana and Schild [64].

All compounds were tested for H_3-antagonistic effects in vitro, following standard methods, using the guinea pig ileum [22].

Male guinea pigs weighing 300–400 g were sacrificed by a blow to the head. A portion of the small intestine, 20–50 cm proximal to the ileocaecal valve (jejunum), was removed and placed in Krebs buffer (composition (mM) NaCl (118), KCl (5.6), $MgSO_4$ (1.18), $CaCl_2$ (2.5), NaH_2PO_4 (1.28), $NaHCO_3$ (25), and glucose (5.5)). Whole jejunum segments (2 cm) were prepared and mounted between two platinum electrodes (4 mm apart) in 20 ml Krebs buffer, continuously gassed with 95% O_2–5% CO_2 and maintained at 37°C. Contractions were recorded isotonically under 1.0 g tension with Hugo Sachs Hebel-Messvorsatz (Tl-2)/HF-modem (Hugo Sacs Electronik, Hugstetten, Germany) connected to a pen recorder. After equilibration for 1 h with washings every 10 min, the muscle segments were stimulated maximally between 15 and 20 V, and continuously at a frequency of 0.1 Hz and a duration of 0.5 ms, with rectangular-wave electrical pulses, delivered by a Grass Stimulator S-88 (Grass Instruments, Quincy, USA). After 30 min of stimulation, cumulative CRCs (half-log increments) of (*R*)-α-methylhistamine were recorded until no change in response was found. The testing compounds were added 20 min before generation of CRCs with (*R*)-α-methylhistamine as H_3-agonist. Between two succeeding measurements, the preparations were washed three times every 10 min, without any stimulation. The data obtained with the described test system are expressed as mean ± SD. Tissue preparations from at least four different animals were used for each compound. Statistical analysis was carried out with the Students *t*-test. In all tests $P < 0.05$ was considered statistically significant. The potency of an antagonist is expressed by its pA_2 value, calculated from the Schild regression analysis where at least three concentrations were used.

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