Synthesis of Novel 4(5)-(5-Aminotetrahydropyran-2-yl)imidazole Derivatives and Their *in Vivo* Release of Neuronal Histamine Measured by Brain Microdialysis

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Received April 27, 2007; accepted May 30, 2007; published online June 1, 2007

The (2*R*,5*S*)-*trans*- and (2*S*,5*S*)-*cis*-stereoisomers 1a and 1b of 4(5)-(5-aminotetrahydropyran-2-yl)imidazole, which have two chiral centers and adopt a stable chair conformation, were synthesized *via* cyclization of diol intermediates 7 using L-glutamine as the starting material. Their enantiomers, (2*S*,5*R*)-*trans*-1c and (2*R*,5*R*)-*cis*-1d, were synthesized by the same methodology from D-glutamine. Stereo isomers 1a—d were converted into cyanoguanidines 11a—d, and into *N*-isopropyl and *N*-3,3-dimethylbutyl derivatives 12a—d and 13a—d, respectively. The results of *in vivo* brain microdialysis of the derivatives apparently indicated that only (2*S*,5*R*)-isomers increased the release of neuronal histamine. Among the many (2*S*,5*R*)-*N*-alkyl derivatives, 13c (OUP-133) and 18 (OUP-153) increased histamine release to 180—190% and 180—200% of basal levels, respectively, and were found to be novel histamine H_3 antagonists.

Key words imidazole; tetrahydropyran; cyclization; H_3; antagonist; microdialysis

Histaminergic neurotransmission in rodent brain is regulated by constitutively active histamine H_3 receptor (H_3R)[1] *in vitro* as well as *in vivo*.[2] H_3R is a presynaptic autoreceptor that is mainly localized in the central nervous system (CNS) and acts to modulate the biosynthesis and release of histamine from histaminergic neurons.[3] H_3R is also a heteroreceptor that modulates the release of a number of neurotransmitters,[4] *e.g.*, dopaminergic[5] and serotonergic[6] systems. H_3-antagonists increase central histamine levels and may therefore be useful for the treatment of a variety of CNS disorders, including eating disorder,[7] schizophrenia,[8] narcolepsy,[9] epilepsy and cognitive disorders, and attention-deficit hyperactivity disorder (ADHD).[10] However, the precise therapeutic potential of H_3-antagoanists remains unknown to this day. After the discovery of the fourth subtype of histamine receptor designated as H_4 receptor (H_4R) in 2000, H_3-antagonists and agonists were found to bind not only to H_3R but also to H_4R.[11—13] In order to investigate the physiological and pathophysiological roles of H_3R and H_4R, the development of more selective H_3R ligands is indispensable.

In studies directed toward the preparation of new H_3 ligands, the cyclopropane ring has been often used as a conformationally restricted spacer located between an imidazole and an amino function, which are essential for the activation of H_3R.[14—16] Indeed, the H_3-antagonist GT-2331 (cipralisant, Perceptin)[17,18] has entered Phase II clinical trials for the treatment of ADHD, and Shuto and co-workers[19] very recently developed potent H_3 antagonists (CAIC) that have the cyclopropane structure (Fig. 1). The introduction of a hydrophobic group to lead compounds also plays a crucial role in the functional inversion of an agonist into the corresponding antagonist. A large number of compounds that were prepared in the course of the search for novel H_3-antagonists have a hydrophobic group on the histamine-related structure, some examples of which are shown in Fig. 1.[20—22] The hydrophobic moiety is usually either an aromatic or a saturated ring system, but it can also be a carbon chain.

We previously examined the effect of four stereoisomers of 4(5)-(5-aminomethyl-tetrahydrofuran-2-yl)imidazole on H_3R in rat brain using *in vivo* microdialysis. Among them, only the (+)-(2*R*,5*R*)-isomer (imifuramine)[23—25] exhibited H_3-agonistic activity that approximately equaled that of immepip (Fig. 2).[26] The tetrahydrofuran (THF) ring of imifuramine may assume flexible conformations by characteristic pseudo-rotations of the five-membered ring. On the other hand, immepip[27] and thioperamide,[3] which are well known as the nonchiral prototypes of H_3R agonists and antagonists, adopt a piperidine spacer having stable chair conformation, and the *N*-function in the piperidine is located four atoms away from the imidazole. With these results and the strategy described previously for the synthesis of new H_3-ligands in mind, we reasoned that as molecular templates, 4(5)-(5-aminotetrahydropyran-2-yl)imidazoles (ATPIs, **1**) might be constrained by a six-membered chair conformation positioned between an imidazole and an amino group, providing four chiral stereoisomers **1a**—**d**, as shown in Fig. 2. Further,

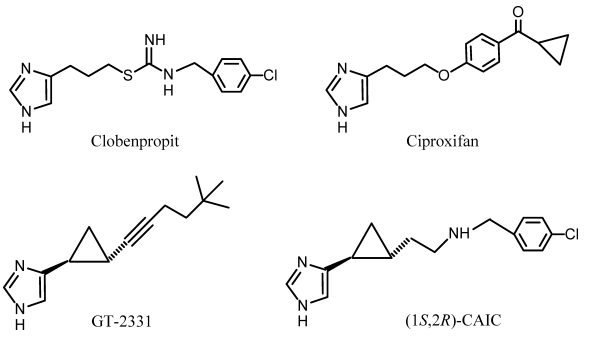

Fig. 1. Structures of Some H_3R Antagonists

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the introduction of various hydrophobic moieties to the amino group in the THP ring may produce many derivatives that would allow us to evaluate their pharmacological effects on H_3R. In the present study, first, we describe the synthesis of novel ATPIs and their *N*-substituted derivatives. Then, their influence on the release of neuronal histamine is investigated using *in vivo* rat brain microdialysis.

Synthesis of ATPIs ATPIs **1a**—**d** were synthesized as shown in Chart 1. Tribenzyl compound **2** were first prepared from L-glutamine with benzyl bromide according to Gmeiner's procedure.[28] Although the optical integrity of **2** were described in the literature,[28] the reaction required 10 days (d) at room temperature (rt) to obtain **2**. We actually obtained **2** in only 42% yield over 13 d in aqueous K_2CO_3 at 40 °C. Microwave (MW) irradiation of organic reactions is known to accelerate a variety of synthetic transformations *via* time- and energy-saving protocols.[29] When we attempted the tribenzylation of L-glutamine under MW irradiation conditions [DMF–H_2O (1 : 1), 80 °C], the reaction time was remarkably shortened to 1 h and the yield of **2** was considerably improved to 69%, making this compound readily available (see Experimental section). Chemoselective reduction (91%) of the ester group of **2** by $LiAlH_4$ at 0 °C to form hydroxy amide **3**, followed by refluxing of **3** in toluene, afforded (5*S*)-5-dibenzylamino-THP-2-one **4** (98%). Reduction of **4** with DIBAL, followed by addition of the lithium salt of bis-protected imidazole **5**[30] to the resulting lactol, gave diol **6** (92%).[31] Hydrolysis of **6** in refluxing 1.5 N HCl afforded diol **7** having an unsubstituted imidazole in 93% yield.

We previously reported an efficient method for the formation of ribofuranosyl *C*-nucleosides or THFs with a five-membered ring *via* a diazafulvene intermediate (like **8**), which involved intramolecular cyclodehydration of the corresponding diols using *N*,*N*,*N*′,*N*′-tetramethylazodicarboxamide (TMAD)[32] and Bu_3P.[31] Therefore, we first attempted treatment of **7** with TMAD (1.5 eq) and Bu_3P (1.5 eq) at rt in benzene for 17 h, followed by *tert*-butoxycarbonylation to facilitate isolation, but N^{im}-Boc-THP intermediate **10** was produced in only 7% yield (Table 1, entry 1). Use of 3 equivalents each of TMAD and Bu_3P under benzene reflux somewhat improved the yield to 43% (entry 2). In contrast, the reaction of diol **6** containing bis-protected imidazole with TMAD-Bu_3P did not proceed at all. These results indicated that cyclization to a six-membered ring using the TMAD-Bu_3P method was more difficult than that to the previous five-membered rings. Alternatively, reagents that activate one hydroxy group to promote the cyclization have been used, such as Burgess reagent,[33] Hendrickson reagent,[34] and Martin's sulfurane.[35] However, the first two reagents did not work at all, and Martin's sulfurane resulted in an inseparable mixture of **10** and diphenylsulfoxide. In an effort to improve the reaction, we turned our attention to the net loss of water using an azeotropic mixture. When diol **7** was refluxed in benzene or toluene in the presence of TsOH (0.1 eq) using a Dean–Stark water separator, the cyclo-dehydration reaction proceeded effectively to afford **10** in 61% or 69% yield (entries 3, 4). The hydrolysis of thus obtained **10** with 1.5 N aqueous HCl, followed by debenzylation afforded 3 : 1 diastereomixture **1**, which was separated easily by silica gel column chromatography to obtain novel chiral (+)-(2*R*,5*S*)-ATPI (**1a**, 75%) and (−)-(2*S*,5*S*)-ATPI (**1b**, 25%) in a *trans* or *cis* relationship. Their enantiomeric units, (−)-(2*S*,5*R*)-*trans*- and (+)-(2*R*,5*R*)-*cis*-ATPIs (**1c**, **1d**), were also obtained from D-glutamine with the present synthetic methodology (Chart 1, Eq. 1).

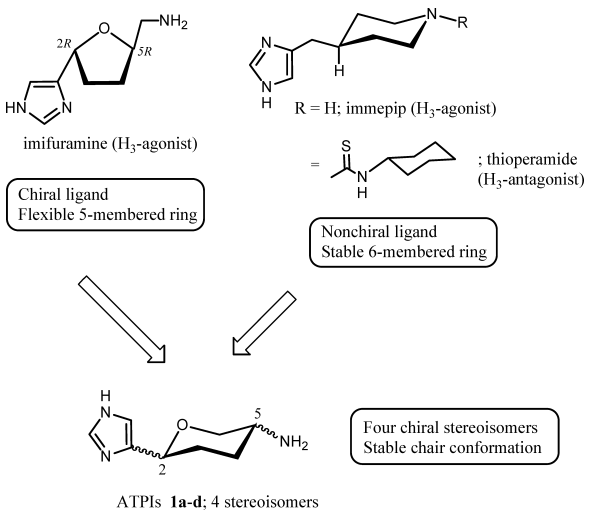
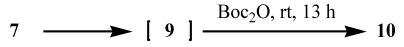


Fig. 2. Chiral 4(5)-(5-Aminotetrahydropyran-2-yl)imidazoles (ATPIs) Having Chair Conformation

The *trans*- or *cis*-stereochemistry of **1a** and **1b** was assigned by detailed observations of ^{1}H-NMR spectra, as illustrated in Fig. 3. *Trans*-isomer **1a** showed ^{1}H–^{1}H NOESY interactions among H-2_{ax}–H-4_{ax}, H-2_{ax}–H-6_{ax}, and H-3_{ax}–H-5_{ax}. In particular, the H-5 resonance at δ=2.80 ppm for **1a** appeared as a triplet of triplets with $J_{5\text{-}6ax}$=$J_{5\text{-}4ax}$=12.4 Hz and $J_{5\text{-}6eq}$=$J_{5\text{-}4eq}$=3.4 Hz. These coupling constants confirmed that H-2 and H-5 occupied axial positions in a chair conformation with the imidazole base and the amino group in equatorial orientations. On the other hand, for **1b**, NOESY data and coupling constant (H-5_{eq}: δ 2.92, broad singlet) indicated *cis*-stereochemistry with the imidazole moiety and the amino group in equatorial and axial orientations, respectively.

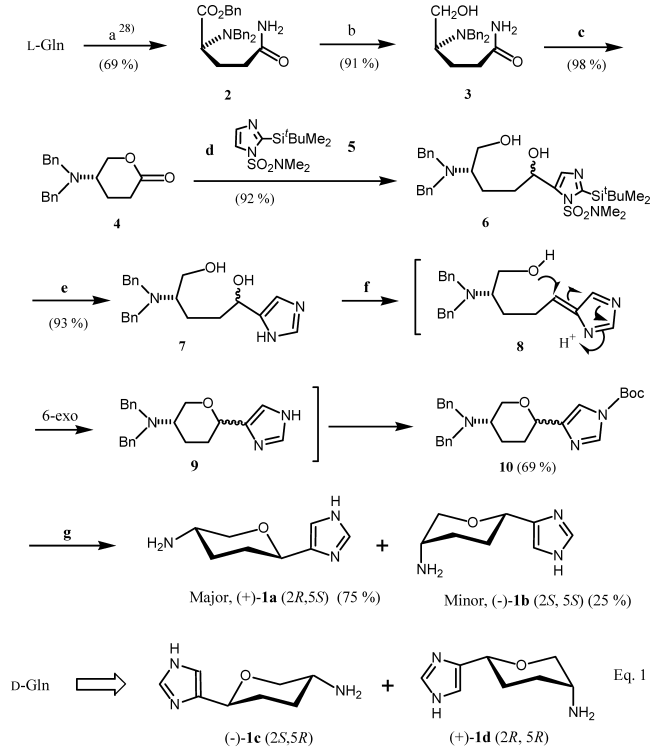
Stereoisomers **1a**—**d** of synthesized ATPI were employed as base compounds in this study. Each isomer was first con-

Table 1. Formation of N^{im}-Boc-THP Intermidiate **10**



Entry	Reagent (eq)	Solvent	Temp.	Time (h)	**10** (%)
1	TMAD (1.5), Bu_3P (1.5)	Benzene–THF	rt	17	7
2	TMAD (3.0), Bu_3P (3.0)	Benzene	Reflux	13	43
3[a]	TsOH (0.1)	Benzene	Reflux	64	61
4[a]	TsOH (0.1)	Toluene	Reflux	24	69

a) Dean–Stark water separator was used.



Reagents and Conditions:(a) Method A (ref. 28): BnBr, K_2CO_3, H_2O, 40 °C, 13 days; Method B: BnBr, K_2CO_3, DMF-H_2O, 80 °C, MW, 1 h; (b) $LiAlH_4$, 0 °C, 40 min; (c) toluene, reflux, 21 h; (d) i) DIBAL, -70 °C, 50 min; ii) lithium salt of **5**, -40 °C then rt, 15 h; (e) 1.5 N HCl; (f) i) TsOH (cat.), reflux, 24 h, toluene, water separator; ii) Boc_2O, Et_3N; (g) i) HCl; ii) H_2 / Pd-C; iii) column chromatography.

Chart 1. Synthesis of ATPIs, **1a**—**d**

verted into three types of ATPI derivatives as shown in Eqs. 1—3 of Chart 2 [The conversions are represented by **1c** (2*S*,5*R*)]. Their reactions provided five types of derivatives, as shown in Table 2; cyanomethylguanidines **11a**—**d** (Table 2, entries 5—8), isopropylamines **12a**—**d** (entries 9—12), 3,3-dimethylbutylamines **13a**—**d** (entries 13—16), (2*R*,5*S*)-

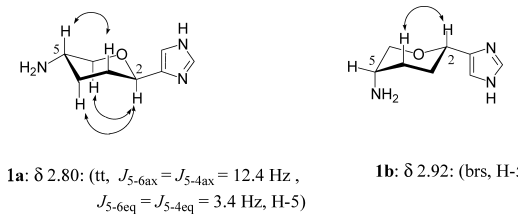


Fig. 3. NOESY Effects and H-5 Coupling Constants for **1a** and **1b**

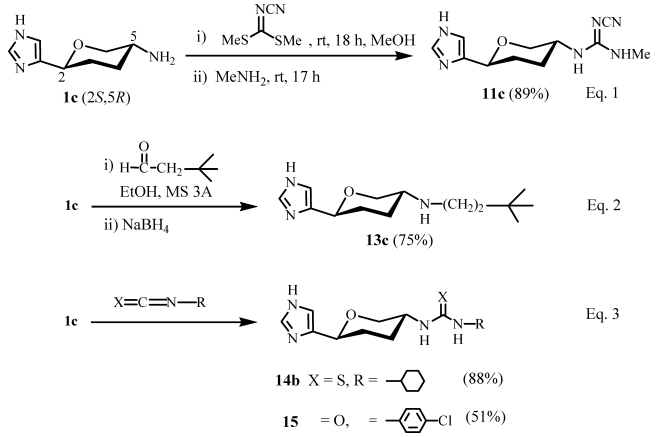
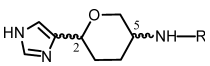
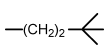
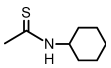
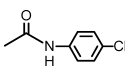


Chart 2. Synthesis of (2*S*,5*R*)-ATPI Derivatives

Table 2. Relationship between Configuration of ATPIs and Neuronal Histamine Release



Entry	Compound	R	Configuration	Histamine release (%)[a,b]
1	**1a**	H	2*R*, 5*S*	N.A.
2	**b**		2*S*, 5*S*	N.A.
3	**c**		2*S*, 5*R*	N.A.
4	**d**		2*R*, 5*R*	N.A.
5	**11a**	NCN ‖ —C—NHMe	2*R*, 5*S*	N.A.
6	**b**		2*S*, 5*S*	N.A.
7	**c**		2*S*, 5*R*	N.A.
8	**d**		2*R*, 5*R*	N.A.
9	**12a**		2*R*, 5*S*	N.A.
10	**b**		2*S*, 5*S*	N.A.
11	**c**		2*S*, 5*R*	+ [120—130%, (N=5)[c]]
12	**d**		2*R*, 5*R*	N.A.
13	**13a**		2*R*, 5*S*	N.A.
14	**b**		2*S*, 5*S*	N.A.
15	**c** (OUP-133)		2*S*, 5*R*	+ [180—190%, (N=5)[c]]
16	**d**		2*R*, 5*R*	N.A.
17	**14a**		2*R*, 5*S*	N.A.
18	**b**		2*S*, 5*R*	+ [120—130%, (N=3)[c]]
19	**15**		2*S*, 5*R*	+ [140—150%, (N=5)[c]]

a) % of basal histamine release. *b*) +: significant increase; N.A.: not active. *c*) N: number of animals.

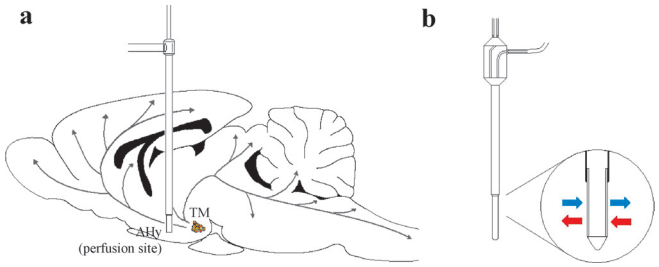


Fig. 4. a) Schematic Model of Histaminergic System and Insertion Area of Microdialysis Probe

The histaminergic system has its cell bodies in the tuberomammillary nucleus (TM) and projects to several brain regions. b) Principle of the microdialysis probe. It can be used to recover and administer substances in living tissue.

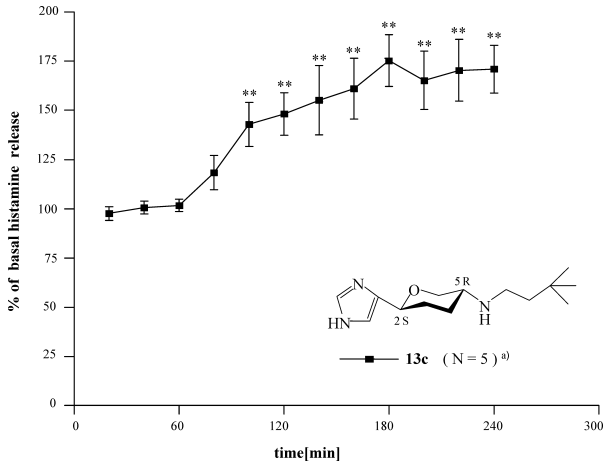


Fig. 5. Effects of **13c** on *in Vivo* Histamine Release in Rat Hypothalamus as Measured by Microdialysis

The compound (10 μM) was infused into the hypothalamus *via* the microdialysis probe. The average value in the first three samples was taken as basal release. Results are expressed as percentages of basal release and are means (±) S.E.M. ∗ Significant (∗ p<0.05, ∗∗ p<0.01) difference between groups was determined by ANOVA with Newman–Keuls proceudre; a) number of animals.

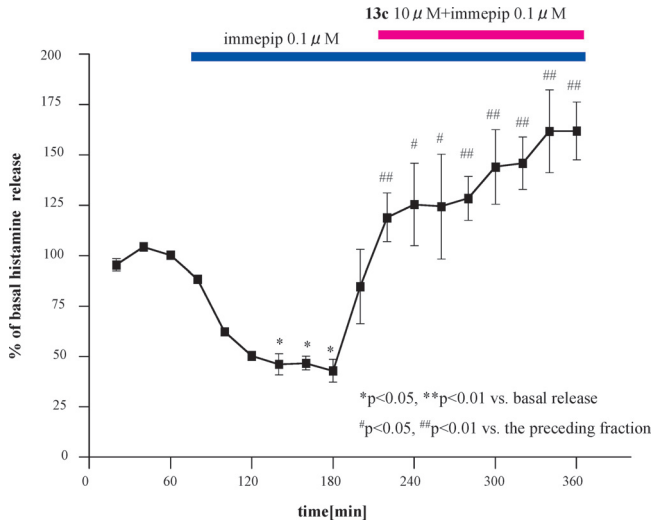


Fig. 6. Effects of **13c** and Immepip on *in Vivo* Histamine Release in Rat Hypothalamus as Measured by Microdialysis

Compounds (**13c**: 10 μM, immepip: 0.01 μM) were infused into the hypothalamus *via* the microdialysis probe. The average value in the first three samples was taken as basal release. Results are expressed as percentages of basal release and are means (±) S.E.M. of four rats. Statistical differences between basal release and the following fractions were detemined by the Newman–Keuls procedure.

and (2*S*,5*R*)-cyclohexylthioureas **14a** and **14b** (entries 17, 18), and (2*S*,5*R*)-4-chlorophenylurea **15** (entry 19).

***In Vivo* Microdialysis Experiments of ATPIs** Pharmacological activities of the four stereoisomers of amino compounds **1a**—**d**, cyanoguanidines **11a**—**d**, isopropylamines **12a**—**d**, and 3,3-dimethylbutylamines **13a**—**d** were first tested by *in vivo* brain microdialysis to examine whether the stereoisomers had any effects on the release of histamine in rat hypothalamus.

In vivo microdialysis is widely used to measure the extracellular levels of many substances in the brain,[36] and is suitable for the determination of neurotransmitter dynamics *in vivo*. A microdialysis probe is implanted into the anterior hypothalamic area (AHy) of rats anesthetized with urethane, as illustrated in Fig. 4a.[37] In all cases, the probe is perfused with artificial cerebrospinal fluid (CSF) containing ATPI derivatives at the concentration of 10 μM, and endogenous histamine in AHy is recovered from the surrounding extracellu-

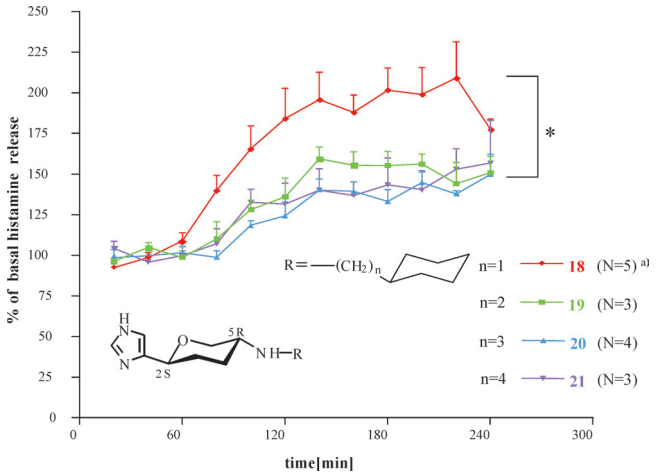


Fig. 7. Effects of **19**, **20**, **21**, and **18** on *in Vivo* Histamine Release in Rat Hypothalamus as Measured by Microdialysis

The experimental and statistical procedures are as described in Fig. 5; a) number of animals.

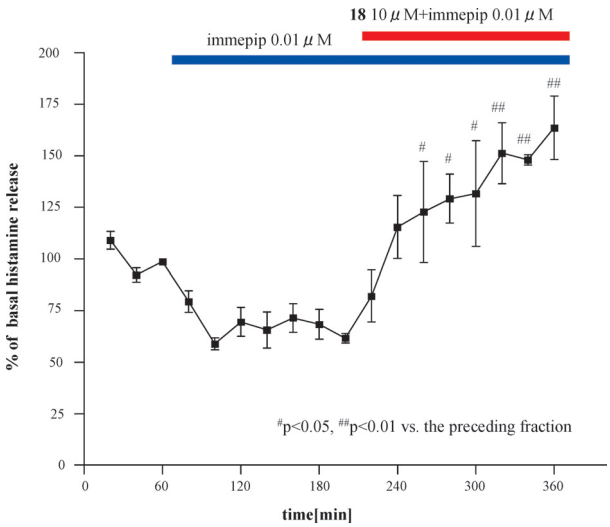


Fig. 8. Effects of **18** and Immepip on *in Vivo* Histamine Release in Rat Hypothalamus as Measured by Microdialysis

Compounds (**18**: 10 μM, immepip: 0.01 μM) were infused into the hypothalamus *via* the microdialysis probe. The experimental and statistical procedures are as described in Fig. 6.

lar fluid through the membrane at the probe tip (Fig. 4b). The fractions are collected every 20 min and histamine analysis is carried out by an HPLC-fluorometric method.[38] The results of evaluation are summarized in Table 2.

Relationship between ATPI Configuration and Neuronal Histamine Release in Rat Brain None of isomers **1a**—**d** and **11a**—**d** had any effects on histamine release (Table 2, entries 1—8). In the case of *N*-isopropylamines **12a**—**d** (entries 9—12), only 2*S*,5*R*-isomer **12c** increased histamine release by approximately 120—130% compared with the basal level (entry 11). In 3,3-dimethylbutylamines **13a**—**d** that contained a bulky *t*-butyl group at the end of the alkyl chain, (2*S*,5*R*)-isomer **13c** showed relatively high histamine release (180—190%; run 15) compared with the basal level, as illustrated in Fig. 5, while other stereoisomers, (2*R*,5*S*)-isomer **13a**, (2*S*,5*S*)-isomer **13b**, and (2*R*,5*R*)-isomer **13d** had no effect on histamine release (Table 2, entries 13, 14, 16).

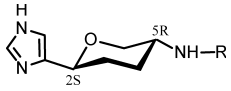
In addition, cyclohexylthiourea **14b** having the 2*S*,5*R*-configuration showed a slight effect on histamine release (120—130%), while enantiomer **14a** had no effect on the release (entries 17, 18). (2*S*,5*R*)-4-Chlorophenylurea **15** moderately increased histamine release by 140—150% compared with the basal level (entry 19). The results of these *in vivo* microdialysis experiments indicated that only the 2*S*,5*R*-configuration of ATPIs induced the release of histamine in rat brain.

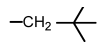
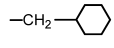
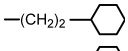
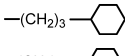
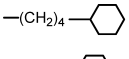
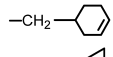
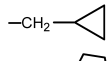
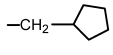
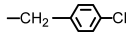
Effect of 13c and Immepip on *in Vivo* Histamine Release The positive findings for (2*S*,5*R*)-*N*-3,3-dimethylbutylamine **13c** motivated us to further investigate the effect of coperfusing **13c** and immepip (Fig. 2),[27] a potent and selective H_3-agonist that has been used in many studies, on *in vivo* histamine release (Fig. 6). The administration of immepip (0.1 μM) alone decreased histamine release to approximately 30% of the basal level, and this effect was apparently reversed by coperfusion of **13c** (10 μM), which increased histamine release to approximately 170% of the basal level. These findings support the idea that the antagonistic activity of **13c** takes place *via* an H_3R.

Influence of (2*S*,5*R*)-*N*-Alkyl ATPIs on Histamine Release in AHy The present H_3-antagonists have three common and essential structural features: the key imidazole head group, a spacer, and a hydrophobic tail group, as shown in Fig. 1. From the results of Table 2, the 2*S*,5*R*-configuration of ATPI derivatives was confirmed to be essential for histamine release. Then, our attention was directed to the optimization of the hydrophobic tail group and the *N*-alkyl chain length. Ten (2*S*,5*R*)-*N*-alkyl-ATPIs **16**—**25** were further prepared by reductive amination of **1c** according to Chart 2, Eq. 2. The yields of the novel *N*-alkyl compounds from **1c** and the results of their microdialysis are summarized in Tables 3 and 4.

Shortening the spacer from an ethylene (**13c**) to a methylene moiety (**16**) reduced somewhat histamine release (130—160%) compared to that of **13c** (Table 4, entry 2). However, cyclohexyl analogue **18**, which has a methylene spacer, gave a high histamine release of 180—200%, the highest in this study (Table 4, entry 4 and Fig. 7). Extended homologues **19**, **20**, and **21**, which have NH and cyclohexane moieties tethered by a long carbon spacer (an ethylene, a propylene, and a butylene moiety, respectively), reduced histamine release (120—160%), as shown in Fig. 7 (Table 4, entries 4—7), while cyclohexyl amine **17**, in which the N atom is directly attached to the cyclohexane ring, was inactive (Table 4, entry 3). Accordingly, among the cyclohexane homologues, the methylene spacer of **18** was optimal for histamine release. In contrast, none of the four hydrophobic moieties: 3-cyclohexenyl, cyclopropyl, cyclopentyl, and 4-chlorophenyl groups (Table 4, entries 8—11) showed activities in spite of the

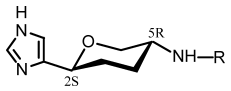
Table 3. *N*-Alkyl Derivatives of (2*S*,5*R*)-ATPI

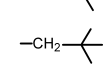
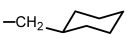
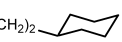
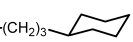
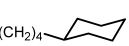
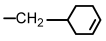
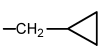
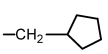
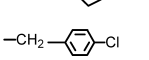


Compound	R	Yield (%)[a]
16		85
17		62
18		74
19		78
20		69
21		60
22		35
23		99
24		96
25		88

a) Yield based on reductive amination of **1c** (Chart 2, Eq. 2).

Table 4. Histamine Release of (2*S*,5*R*)-*N*-Alkyl-ATPIs



Entry	Compound	R	Histamine release (%)[a,b]
1	**13c** (OUP-133)		+ [180—190 (N=5)[c]]
2	**16**		+ [130—160 (N=4)[c]]
3	**17**		N.A.
4	**18** (OUP-153)		+ [180—200 (N=5)[c]]
5	**19**		+ [150—160 (N=3)[c]]
6	**20**		+ [120—150 (N=4)[c]]
7	**21**		+ [130—150 (N=3)[c]]
8	**22**		N.A.
9	**23**		N.A.
10	**24**		N.A.
11	**25**		N.A.

a) % of basal histamine release. *b*) +: significant increase; N.A.: not active. *c*) N: number of animals.

methylene spacer. The results suggested that the moieties that are almost to flat or planar, were unsuitable as the hydrophobic tail group of ATPIs.

To clarify whether **18** is an H_3-antagonist, we again examined its antagonistic activity against immepip[27] (Fig. 8). The administration of immepip (0.01 μM) to the perfusion fluid decreased histamine release to approximately 60% of the basal level. Co-infusion of **18** (10 μM) fully antagonized this effect and increased histamine release to approximately 160% of the basal level. This indicated that the H_3-antagonistic activity of **18** was mediated by H_3R.

Conclusion

Novel ATPI isomers **1a**—**d** were synthesized from L- or D-glutamine (Chart 1). Then, five types of *N*-substituted derivatives based on **1a**—**d** were prepared to examine their influence on histamine release in rat brain through *in vivo* brain microdialysis experiments (Chart 2). The results are summarized as follows: 1) Basal amino isomers **1a**—**d** are inactive, although some of the *N*-substituted derivatives enhance histamine release (Table 2). 2) The 2*S*,5*R*-configuration of the THP ring is the bioactive configuration for histamine release. 3) *N*-3,3-Dimethylbutylamine **13c** (OUP-133) and cyclohexylmethylamine **18** (OUP-153) increase histamine release to 180—190% and 180—200% of their basal levels, respectively (Tables 2, 4). 4) Their antagonistic activities against immepip indicate that **13c** and **18** are H_3-antagonists (Figs. 6, 8). 5) The carbon chain length (*n*) between NH and a hydrophobic tail group of $n=1$ or 2 is appropriate, and a bulky substituent (*tert*-butyl or cyclohexyl) is preferred to a plane-like substituent as the hydrophobic tail group (Table 4, entries 8—11). 6) Previous studies have shown that imifuramine derivatives having flexible five-membered THF structures generally exhibit H_3-agonistic activities.[23,25] In contrast, this study has shown that six-membered ATPIs having stable chair spacers exhibit only H_3-antagonistic activities. Armed with the findings of H_3-antagonists **13c** and **18**, the synthesis of related ATPI derivatives and studies of their pharmacological effects are in progress in our laboratories.

Experimental

General Melting points were determined on a hot-stage apparatus and were uncorrected. Optical rotation measurements were recorded with a JASCO DIP-1000 digital polarimeter. IR spectra were recorded on a Shimadzu IR-435 spectrometer. ^{1}H- and ^{13}C-NMR spectra were measured with tetramethylsilane as internal standard on Varian Gemini-200, Varian Mercury-300, and Varian UNITY INOVA-500 spectrometers. Reactions with air- and moisture-sensitive compounds were carried out under argon atmosphere. Unless otherwise noted, all extracts were dried over Na_2SO_4 and the solvent was removed in a rotary evaporator under reduced pressure. BW-127ZH (Fuji Silysia Chemical Ltd.) and Chromatorex NH-DM 1020 [(NH-silica gel), Fuji Silysia Chemical Ltd.] were used for column chromatography. Dehydrated THF was purchased from Wako Pure Chemical Industries, Ltd. TLC was performed on pre-coated TLC plates with $60F_{254}$ (silica gel, Merck). MW-assisted reaction of the first step was performed in a Milestone MicroSYNTH multimodal reactor with thermal control.

(2*S*)-Benzyl 4-Carbamoyl-2-dibenzylaminobutyrate (2). Method A (Gmeiner's Procedure)[28] To a solution of L-glutamine (10.2 g, 69.9 mmol) and K_2CO_3 (47.7 g, 343 mmol) in H_2O (400 ml) was added BnBr (34 ml, 280 mmol). The mixture was well stirred with a mechanical stirrer for 13 d while maintaining the temperature at 40 °C. After the reaction, the reaction mixture was extracted twice with ethyl acetate and dried. The organic layer was evaporated to give a residue that was purified by silica gel (BW-127ZH) column chromatography [*Rf* (50% ethyl acetate/hexane); 0.19]. Elution with 20 to 100% ethyl acetate afforded **2** (12.24 g, 42%) as a pale yellow oil (Gmeiner and co-worker[28] obtained **2** in 46% after stirring 10 d at rt).

Method B In a 30 ml Teflon MW reaction vessel, K_2CO_3 (3.38 g, 24.5 mmol) in H_2O (8 ml) was added to a solution of L-glutamine (730 mg, 5 mmol) in DMF (10 ml). Then, BnBr (2.4 ml, 20 mmol) was added and the vessel was sealed and heated in the Milestone MicroSYNTH MW reactor to 80 °C over a 10 min period. The reaction was held at this temperature for 50 min and was allowed to cool thereafter. The contents were diluted with H_2O and ethyl acetate. The organic layer was separated, dried, and evaporated to give a residue that was then subjected to chromatography (BW-127ZH). Elution with 20 to 80% ethyl acetate/hexane afforded **2** (1.44 g, 69%) as a pale yellow oil. ^{1}H-NMR ($CDCl_3$) δ: 1.95—2.20 (2H, m), 2.21—2.35 (2H, m), 3.29—3.43 (1H, m), 3.50 (2H, d, J=21.6 Hz), 3.87 (2H, d, J=18.0 Hz), 4.94—5.10 (2H, m), 5.13—5.32 (2H, m), 7.12—7.49 (15H, m). (The $[\alpha]_D$ of ATPI **1a** derived from **2** prepared by Method B showed the same value as that of Method A.)

(4*S*)-4-Dibenzylamino-5-hydroxypentanamide (3) To a suspension of lithium aluminum hydride (2.23 g, 58.6 mmol) in ether (100 ml) was added dropwise a solution of **2** (12.18 g, 29.3 mmol) in ether (120 ml) at 0 °C, and the mixture was stirred for 40 min at the same temperature. Then, water (10 ml), 15% aqueous sodium hydroxide (10 ml), and water (10 ml) were successively added to the reaction mixture. The resultant mixture was dried over anhydrous magnesium sulfate, filtered through Celite, and evaporated. The residue was purified by silica gel (BW-127ZH) column chromatography (10% methanol/ethyl acetate) to afford **3** (8.30 g, 91%) as a pale yellow oil. ^{1}H-NMR ($CDCl_3$) δ: 1.51—1.78 (2H, m), 2.10—2.23 (2H, m), 2.68—2.82 (1H, m), 2.97—3.08 (1H, br), 3.45—3.58 (4H, m), 3.80 (2H, d, J=13.1 Hz), 5.30—5.47 (2H, br), 7.25—7.37 (10H, m). EI-MS: *m*/*z*: 313 (M^++1). HR-MS: *m*/*z*: 313.1918 (Calcd for $C_{19}H_{25}N_2O_2$: 313.1915).

(5*S*)-5-Dibenzylaminotetrahydropyran-2-one (4) A solution of **3** (8.25 g, 26.4 mmol) in toluene (570 ml) was refluxed for 21 h. After evaporation, the residue was purified by silica gel (BW-127ZH) column chromatography (50% ethyl acetate/hexane) to afford **4** (7.66 g, 98%) as a pale yellow oil. IR (film) cm^{-1}: 1730. ^{1}H-NMR ($CDCl_3$) δ: 1.85—2.20 (2H, m), 2.32—2.53 (1H, m), 2.57—2.74 (1H, m), 3.17—3.32 (1H, m), 3.59—3.78 (4H, m), 4.29—4.34 (2H, m), 7.23—7.38 (10H, m). EI-MS *m*/*z*: 295 (M). HR-MS *m*/*z*: 295.1571 (Calcd for $C_{19}H_{21}NO_2$: 295.1571).

2-*tert*-Butyldimethylsilyl-5-[(1*RS*,4*S*)-4-dibenzylamino-1,5-dihydroxypentyl]imidazole-*N*,*N*-dimethylsulfonamide (6) To a solution of **4** (6.67 g, 22.6 mmol) in dry toluene (120 ml) at −70 °C was added dropwise a 1 M solution of DIBAL in toluene (45.2 ml, 45.2 mmol) over 15 min. After stirring for 50 min at −70 °C, the reaction mixture was quenched with methanol (40 ml) and further stirred for 30 min at rt. Saturated sodium bicarbonate (40 ml) solution was added and the reaction mixture was stirred for another 10 min. After anhydrous magnesium sulfate was added to the resulting suspension, the reaction mixture was stirred for 30 min, filtered through a Celite pad, and washed with ethyl acetate. The filtrate was evaporated to give a crude oil (5-dibenzylaminotetrapyran-2-ol). Alternatively, a solution of **5** (19.6 g, 67.8 mmol) in THF (66 ml) was cooled to −40 °C, and 1.6 M BuLi-hexane (42 ml, 67.8 mmol) was added dropwise over 15 min to precipitate white lithium salt of **5**. The resulting suspension was stirred for 15 min and cooled to −30 °C. Then, a toluene solution (45 ml) of 5-dibenzylaminotetrapyran-2-ol prepared above was added slowly over 20 min at the same temperature, and the whole mixture was stirred for 5 min. The dry ice bath was removed and the reaction mixture was stirred at rt to dissolve the salts. After 15 h, a small amount of water was added to the mixture and the solvent was evaporated to give a residue that was extracted twice with ethyl acetate. The extract was washed with water and brine, dried, and evaporated to give a crude oil, which was purified by column chromatography (BW-127ZH, 50% ethyl acetate/hexane) to give **6** (12.12 g, 92%) as amorphous powder. ^{1}H-NMR ($CDCl_3$) δ: 0.45 (6H, s), 1.02 (9H, s), 1.21—1.38 (1H, m), 1.78—2.08 (3H, m), 2.76—3.20 (9H, m), 3.42—3.59 (4H, m), 3.80 (2H, m), 4.79—4.89 (1H, m), 7.15—7.42 (11H, m). SI-MS *m*/*z*: 587 (M^++1). HR-MS *m*/*z*: 587.3077 (Calcd for $C_{30}H_{47}N_4O_4SSi$: 587.3085).

4(5)-[(1*RS*,4*S*)-4-Dibenzylamino-1,5-dihydroxypentyl]imidazole (7) To a THF solution (4 ml) of **6** (425 mg, 0.725 mmol) was added 1.5 N HCl (6 ml). The resulting mixture was refluxed for 3.5 h and neutralized by adding 30% NH_4OH. The solvent was evaporated to give a residue that was extracted with ethyl acetate three times by the salting-out technique. The combined organic layer was dried and evaporated to give a crude oil that was subjected to column chromatography (BW-127ZH) using 20% methanol/ethyl acetate as eluent to give **7** (247 mg, 93%) as an amorphous product. ^{1}H-NMR ($CDCl_3$) δ: 1.18—1.30 (1H, m), 1.62—1.95 (3H, m),

2.61—2.79 (1H, m), 3.36—3.51 (4H, m), 3.63—3.75 (2H, m), 4.58 (1H, t, J=9.2 Hz), 6.10—6.61 (2H, br), 6.78 (1H, s), 7.12—7.36 (11H, m). SI-MS m/z: 366 (M^++1). HR-MS m/z: 366.2182 (Calcd for $C_{22}H_{28}N_3O_2$: 366.2180).

***tert*-Butyl-4-[(2*RS*,5*S*)-5-dibenzylaminotetrahydropyran-2-yl]imidazole-1-carboxylate (10)** The solution of **7** (130 mg, 0.356 mmol) in toluene (50 ml) containing a catalytic amount TsOH (6 mg, 0.036 mmol) was refluxed for 24 h as an azeotrope using a Dean-Stark water separator (molecular sieve 4A). After evaporation followed by neutralization by the addition of 30% NH_4OH, the resulting mixture was extracted three times with chloroform, dried over anhydrous magnesium sulfate, and evaporated to give a crude oil of **9**. A solution of Boc_2O (155 mg, 0.71 mmol) in THF (1.5 ml) and then triethylamine (0.10 ml, 0.71 mmol) were added to the solution of **9** in THF (1.5 ml), and the resulting mixture was stirred at rt for 17 h. The solvent was evaporated. Chromatography on silica gel using 20% ethyl acetate/hexane as eluent gave **10** (109 mg, 69%) as a colorless oil. ^{1}H-NMR ($CDCl_3$) δ: 1.45—2.27 (13H, m), 2.76—2.94 (1H, m), 3.52—3.93 (5H, m), 4.07—4.16 (1H, m), 4.24—4.32 (3/4H, m), 4.65—4.70 (1/4H, m), 7.14—7.43 (11H, m), 7.98 (3/4H, s), 8.02 (1/4H, s). SI-MS m/z: 448 (M^++1). HR-MS m/z: 448.2600 (Calcd for $C_{27}H_{34}N_3O_3$: 448.2598).

4(5)-[(2*R*,5*S*)-5-Aminotetrahydropyran-2-yl]imidazole [(+)-1a] and 4(5)-[(2*S*,5*S*)-5-Aminotetrahydropyran-2-yl]imidazole [(−)-1b] To a solution of **10** (2.27 g, 5.08 mmol) in ethanol (40 ml) was added 1 N HCl (25 ml). The resulting mixture was stirred for 2 h at rt and the solvent was evaporated to give a residual oil. The oil was further diluted with ethanol and evaporated to remove water as an azeotrope. The operation was carried out again to give a pale yellow oil of **9** dihydrochloride. A solution of **9** dihydrochloride in EtOH (45 ml) was subsequently hydrogenated on 10% Pd–C (2.9 g) at an initial pressure of 3.0 kg/cm^2 for 19 h. The catalyst was removed by filtration through filter paper, and the filtrate was evaporated to give a residue (**1**·2HCl). The dihydrochloride was subjected to column chromatography (NH-silica gel) using $CHCl_3$–MeOH–30%NH_4OH (75 : 25 : 3). The eluents gave (+)-**1a** (640 mg, 75%) and (−)-**1b** (208 mg, 25%) as colorless oils in succession.

(+)-**1a**: $[\alpha]_D$=+17.1° (c=3.1, MeOH). ^{1}H-NMR (CD_3OD) δ: 1.42 (1H, qd, J=12.4, 3.4 Hz, 3′-H_{ax}), 1.82 (1H, qd, J=12.4, 3.4 Hz, 2′-H_{ax}), 1.91—2.03 (1H, m, 2′-H_{eq}), 2.03—2.17 (1H, m, 3′-H_{eq}), 2.80 (1H, tt, J=12.4, 3.4 Hz, 4′-H), 3.20 (1H, dd, J=12.4, 3.4 Hz, 5′-H_{ax}), 3.98 (1H, ddd, J=15.2, 6.1, 2.8 Hz, 5′-H_{eq}), 4.33 (1H, dd, J=12.4, 12.4 Hz, 1′-H), 6.99 (1H, s, 5-H), 7.63 (1H, s, 2-H). ^{13}C-NMR (CD_3OD) δ: 31.6, 33.2, 48.3 (overlapped with CH_3OH in CD_3OD), 74.2, 74.5, 117.2, 136.4, 139.8. EI-MS m/z: 167 (M^+). HR-MS m/z: 167.1061 (Calcd for $C_8H_{13}N_3O$: 167.1058). (−)-**1b**: $[\alpha]_D$=−31.6° (c=2.9, MeOH). ^{1}H-NMR (CD_3OD) δ: 1.66—1.78 (1H, m, 2′-H_{eq}), 1.85—1.94 (1H, m, 3′-H_{eq}), 1.98—2.05 (1H, m, 3′-H_{ax}), 2.05—2.21 (1H, m, 2′-H_{ax}), 2.92 (1H, br s, 4′-H), 3.77 (2H, s, 5′-H), 4.46 (1H, dd, J=10.4, 2.3 Hz, 1′-H), 7.00 (1H, s, 5-H), 7.63 (1H, s, 2-H). ^{13}C-NMR (CD_3OD) δ: 27.4, 31.6, 48.6 (overlapped with CH_3OH in CD_3OD), 73.7, 74.8, 117.8, 135.0, 139.6. EI-MS: m/z=167 (M^+). HR-MS m/z: 167.1055 (Calcd for $C_8H_{13}N_3O$: 167.1058).

Configuration counterparts (−)-**1c** and (+)-**1d** were synthesized with the present method from D-glutamine: (−)-**1c**: $[\alpha]_D$=−14.7° (c=2.7, MeOH); (+)-**1d**: $[\alpha]_D$=+22.2° (c=2.7, MeOH).

2-Cyano-1-methyl-3-{(2*S*,5*R*)-2-[1*H*-imidazol-4(5)-yl]tetrahydropyran-5-yl}guanidine (11c) A solution of **1c** (30 mg, 0.180 mmol) and dimethyl *N*-cyanodithioiminocarbonate (58 mg, 0.36 mmol) was stirred at rt for 18 h, and then 40% $MeNH_2$ in MeOH (4 ml) was added. The resulting mixture was stirred for 17 h at rt. The solvent was evaporated to give a residual oil that was chromatographed [NH-silica gel, MeOH–AcOEt (1 : 19)] to give **11c** (40 mg, 89%) as colorless oil. $[\alpha]_D$=−19.6° (c=3.7, MeOH). IR (film) cm^{-1}: 2160 (CN). ^{1}H-NMR (CD_3OD) δ: 1.58—2.18 (4H, m), 2.79 (3H, s), 3.26—3.35 (2H, overlapped with CH_3OH in CD_3OD, 1H, 5′-H), 3.74—3.91 (1H, m), 4.03 (1H, ddd, J=15.2, 6.1, 2.8 Hz, 5′-H), 4.38 (1H, dd, J=15.2, 3.4 Hz), 7.00 (1H, s), 7.62 (1H, s). ^{13}C-NMR (CD_3OD) δ: 29.0, 30.8, 31.8, 49.0 (overlapped with CH_3OH in CD_3OD), 71.4, 74.4, 116.5, 119.6, 136.2, 139.1, 160.8. EI-MS m/z: 248 (M^+). HR-MS m/z: 248.1382 (Calcd for $C_{11}H_{16}N_6O$: 248.1385).

2-Cyano-1-methyl-3-{(2*R*,5*R*)-2-[1*H*-imidazol-4(5)-yl]tetrahydropyran-5-yl}guanidine (11d) Using the same procedure as that for the preparation of **11c**, **1d** (27 mg, 0.162 mmol) was converted into **11d** (35 mg, 88%) as a colorless oil. $[\alpha]_D$=+22.8° (c=4.0, MeOH). IR (film) cm^{-1}: 2160 (CN). ^{1}H-NMR (CD_3OD) δ: 1.74—2.13 (4H, m), 2.94 (3H, s), 3.77—3.93 (3H, m), 4.51 (1H, dd, J=10.5, 2.1 Hz), 7.02 (1H, s), 7.62 (1H, s). ^{13}C-NMR (CD_3OD) δ: 27.4, 28.5, 28.9, 47.7, 71.0, 74.3, 116.2, 119.4, 135.8, 139.4, 160.5. EI-MS m/z: 248 (M^+). HR-MS: m/z: 248.1386 (Calcd for $C_{11}H_{16}N_6O$: 248.1385).

4(5)-[(2*S*,5*R*)-5-(3,3-Dimethylbutylamino)tetrahydropyran-2-yl]-1*H*-imidazole (13c: OUP-133); General Procedure for the Preparation of *N*-Alkyl Derivatives 12a—d, 13a, 13b, 13d, and 16—25 To a solution of **1c** (46 mg, 0.275 mmol) and 3,3-dimethylbutylaldehyde (0.35 ml, 2.8 mmol) in 4 ml of absolute ethanol was added 3A molecular sieves (220 mg). After stirring the mixture at rt for 5 h, sodium borohydride (213 mg, 5.6 mmol) was added and the mixture was stirred for 23 h at rt. Then, the reaction mixture was diluted with ethanol (5 ml) and neutralized with 2 M acetic acid. The resulting insoluble material was filtered through Celite pad, and the filtrate was evaporated to give a residue that was purified by column chromatography using $CHCl_3$–MeOH–30%NH_4OH (85 : 15 : 1) to give colorless oil **13c** (52 mg, 75%). $[\alpha]_D$=−18.3° (c=4.1, MeOH). ^{1}H-NMR (CD_3OD) δ: 0.93 (9H, s), 1.34—1.50 (3H, m), 1.76—1.91 (1H, m), 1.95—2.04 (1H, m), 2.13—2.24 (1H, m), 2.54—2.76 (3H, m), 3.21—3.33 (1H, overlapped with CH_3OH in CD_3OD, 5′-H), 4.10 (1H, ddd, J=10.9, 4.7, 2.4 Hz), 4.35 (1H, dd, J=11.8, 2.2 Hz), 6.97 (1H, s), 7.58 (1H, s). ^{13}C-NMR (CD_3OD) δ: 30.1, 30.7, 31.3, 31.6, 43.9, 44.6, 54.7, 72.6, 74.7, 116.8, 135.5, 139.3. EI-MS m/z: 252 (M^++1). HR-MS m/z: 252.2078 (Calcd for $C_{14}H_{26}N_3O$: 252.2074).

4(5)-[(2*R*,5*R*)-5-(3,3-Dimethylbutylamino)tetrahydropyran-2-yl]-1*H*-imidazole (13d) Colorless oil. $[\alpha]_D$=+28.4° (c=3.7, MeOH). ^{1}H-NMR (CD_3OD) δ: 0.96 (9H, s), 1.44—1.55 (2H, m), 1.65—1.76 (1H, m), 1.79—1.94 (1H, m), 1.96—2.11 (2H, m), 2.60—2.78 (3H, m), 3.70 (1H, dd, J=12.3, 2.7 Hz), 3.95 (1H, ddd, J=12.3, 2.7, 2.5 Hz), 4.48 (1H, dd, J=10.2, 3.8 Hz), 7.00 (1H, s), 7.62 (1H, s). ^{13}C-NMR (CD_3OD) δ: 27.4, 27.6, 30.1, 30.7, 43.7, 44.1, 52.6, 70.2, 74.2, 117.4, 135.7, 138.9. EI-MS m/z: 251 (M^+). HR-MS m/z: 251.1991 (Calcd for $C_{14}H_{25}N_3O$: 251.1996).

4(5)-[(2*S*,5*R*)-5-Isopropylaminotetrahydropyran-2-yl]-1*H*-imidazole (12c) Colorless oil. $[\alpha]_D$=−21.8° (c=3.7, MeOH). ^{1}H-NMR (CD_3OD) δ: 1.06 (3H, d), 1.09 (d, 3H, J=6.4 Hz), 1.34—1.49 (1H, m), 1.76—1.91 (1H, m), 1.93—2.04 (1H, m), 2.11—2.22 (1H, m), 2.81 (1H, dddd, J=10.9, 10.9, 4.6, 4.5 Hz), 2.99 (1H, heptet, J=6.4 Hz), 3.25 (1H, dd, J=10.9, 10.8 Hz), 4.07 (1H, ddd, J=10.9, 4.6, 2.5 Hz), 4.35 (1H, dd, J=11.1, 2.5 Hz), 6.97 (1H, s), 7.59 (1H, s). ^{13}C-NMR (CD_3OD) δ: 22.6, 31.1, 31.6, 46.5, 51.3, 72.4, 74.7, 116.7, 135.6, 139.1. EI-MS m/z: 209 (M^+). HR-MS m/z: 209.1530 (Calcd for $C_{11}H_{19}N_3O$: 209.1527).

4(5)-[(2*R*,5*R*)-5-Isopropylaminotetrahydropyran-2-yl]-1*H*-imidazole (12d) Colorless oil. $[\alpha]_D$=+30.9° (c=1.9, MeOH). ^{1}H-NMR (CD_3OD) δ: 1.08 (3H, d, J=6.7 Hz), 1.11 (3H, d, J=6.7 Hz), 1.65—2.09 (4H, m), 2.78—2.84 (1H, m), 3.00 (1H, heptet, J=6.7 Hz), 3.72 (1H, dd, J=11.8, 2.4 Hz), 3.90 (1H, ddd, J=11.8, 2.6, 2.5 Hz), 4.48 (1H, dd, J=10.7, 2.8 Hz), 7.00 (1H, s), 7.62 (1H, s). ^{13}C-NMR (CD_3OD) δ: 22.3, 22.9, 27.3, 49.1, 70.9, 74.3, 117.2, 135.8, 139.1. EI-MS m/z: 209 (M^+). HR-MS m/z: 209.1533 (Calcd for $C_{11}H_{19}N_3O$: 209.1527).

4(5)-[(2*S*,5*R*)-5-(2,2-Dimethylpropylamino)tetrahydropyran-2-yl]-1*H*-imidazole (16) Oil. ^{1}H-NMR (CD_3OD) δ: 0.95 (9H, s), 1.25—1.48 (1H, m), 1.74—1.90 (1H, m), 1.94—2.04 (1H, m), 2.10—2.24 (1H, m), 2.34—2.70 (3H, m), 3.21 (1H, dd, J=12.3, 2.7 Hz), 4.10 (1H, ddd, J=12.3, 2.7, 2.5 Hz), 4.35 (1H, dd, J=10.2, 3.8 Hz), 6.98 (1H, s), 7.60 (1H, s).

4(5)-[(2*S*,5*R*)-5-Cyclohexylaminotetrahydropyran-2-yl]-1*H*-imidazole (17) Oil. $[\alpha]_D$=+21.1° (c=0.82, EtOH). ^{1}H-NMR (CD_3OD) δ: 0.98—1.48 (6H, m), 1.60—2.04 (7H, m), 2.10—2.20 (1H, m), 2.52—2.63 (1H, m), 2.81—2.91 (1H, m), 3.2 (1H, overlapped with CH_3OH in CD_3OD), 4.06 (1H, ddd, J=7.5, 2.9, 1.5 Hz), 4.35 (1H, dd, J=7.5, 1.5 Hz), 6.97 (1H, s). EI-MS m/z: 249 (M^+). HR-MS m/z: 249.1840 (Calcd for $C_{14}H_{23}N_3O$: 249.1840).

4(5)-[(2*S*,5*R*)-5-Cyclohexylmethylaminotetrahydropyran-2-yl]-1*H*-imidazole (18: OUP-153) White powder. $[\alpha]_D$=−13.5° (c=1.5, MeOH). ^{1}H-NMR (CD_3OD) δ: 0.8—1.80 (12H, m), 1.80—1.89 (1H, m), 1.89—2.08 (1H, m), 2.08—2.27 (1H, m), 2.40—2.52 (2H, dd, J=9.3, 5.1 Hz), 2.56—2.73 (1H, m), 3.21—3.27 (1H, m), 4.05—4.15 (1H, ddd, J=11.0, 4.5, 3.0 Hz), 4.30—4.41 (1H, dd, J=11.0, 3.0 Hz), 6.99 (1H, s), 7.61 (1H, s). SI-MS m/z: 264 (M^++1). HR-MS m/z: 264.2073 (Calcd for $C_{15}H_{26}N_3O$: 264.2074).

4(5)-[(2*S*,5*R*)-5-Cyclohexylethylaminotetrahydropyran-2-yl]-1*H*-imidazole (19) Oil. $[\alpha]_D$=−14.8° (c=2.0, EtOH). ^{1}H-NMR (CD_3OD) δ: 0.86—2.22 (15H, m), 2.66—2.76 (3H, m), 3.25 (2H, t, J=10.9 Hz), 4.10 (1H, ddd, J=10.9, 4.2, 2.4 Hz), 4.35 (1H, dd, J=11.5, 2.4 Hz), 6.96 (1H, s), 7.59 (1H, d, J=1.1 Hz). EI-MS m/z: 278 (M^++1). HR-MS m/z: 278.2229 (Calcd for $C_{16}H_{28}N_3O$: 278.2230).

4(5)-[(2*S*,5*R*)-5-Cyclohexylpropylaminotetrahydropyran-2-yl]-1*H*-imidazole (20) Oil. $[\alpha]_D$=−7.0° (c=2.1, EtOH). ^{1}H-NMR (CD_3OD) δ: 0.86—1.76 (16H, m), 1.80—1.90 (1H, m), 1.94—2.04 (1H, m), 2.13—2.22 (1H, m), 2.52—2.64 (2H, m), 2.66—2.75 (1H, m), 3.49—3.68 (1H, m), 4.10

(1H, ddd, J=8.9, 2.6, 1.3 Hz), 4.36 (1H, dd, J=10.6, 1.3 Hz), 6.98 (1H, s), 7.60 (1H, s). EI-MS m/z: 292 (M^++1). HR-MS m/z: 291.2303 (Calcd for $C_{17}H_{29}N_3O$: 292.2390).

4(5)-[(2*S*,5*R*)-5-Cyclohexylbutylaminotetrahydropyran-2-yl]-1*H*-imidazole (21) Oil. $[\alpha]_D$=−13.9° (c=1.9, EtOH). ^{1}H-NMR (CD_3OD) δ: 0.82—2.24 (21H, m), 2.56—2.67 (2H, m), 2.68—2.77 (1H, m), 3.17—3.35 (1H, m), 4.10 (1H, ddd, J=10.0, 4.5, 2.6 Hz), 4.36 (1H, dd, J=10.6, 2.6 Hz), 6.98 (1H, s), 7.59 (1H, d, J=1.3 Hz). EI-MS m/z: 305 (M^+). HR-MS m/z: 305.2467 (Calcd for $C_{18}H_{31}N_3O$: 305.2470).

4(5)-[(2*S*,5*R*)-5-(Cyclohex-3-enylmethylamino)tetrahydropyran-2-yl]-1*H*-imidazole (22) Oil. $[\alpha]_D$=−12.5° (c=1.0, EtOH). ^{1}H-NMR (CD_3OD) δ: 1.14—2.23 (11H, m), 2.56 (2H, dd, J=5.8, 3.8 Hz), 2.61—2.78 (1H, m), 3.22—3.34 (1H, m), 4.12 (1H, ddd, J=11.0, 4.4, 2.2 Hz), 4.36 (1H, dd, J=11.0, 2.6 Hz), 5.64 (2H, d, J=2.3 Hz), 6.97 (1H, s), 7.59 (1H, s). EI-MS m/z: 261 (M^+). HR-MS m/z: 261.1835 (Calcd for $C_{15}H_{23}N_8O$: 261.1840).

4(5)-[(2*S*,5*R*)-5-Cyclopropylmethylaminotetrahydropyran-2-yl]-1*H*-imidazole (23) Oil. ^{1}H-NMR (CD_3OD) δ: 0.16—1.02 (5H, m), 0.81—1.52 (4H, m), 1.67—1.90 (1H, m), 2.12—2.23 (1H, m), 2.46—2.80 (3H, m), 3.28 (1H, dd, J=12.3, 2.7 Hz), 4.08 (1H, ddd, J=12.3, 2.7, 2.7 Hz), 4.35 (1H, dd, J=10.2, 3.8 Hz), 6.98 (1H, s), 7.59 (1H, s).

4(5)-[(2*S*,5*R*)-5-Cyclopentylmethylaminotetrahydropyran-2-yl]-1*H*-imidazole (24) Oil. ^{1}H-NMR (CD_3OD) δ: 0.82—2.21 (14H, m), 2.46—2.77 (3H, m), 3.20 (1H, dd, J=12.3, 2.7 Hz), 4.10 (1H, ddd, J=12.3, 2.7, 2.5 Hz), 4.35 (1H, dd, J=10.2, 3.8 Hz), 6.98 (1H, s), 7.59 (1H, s).

4(5)-[(2*S*,5*R*)-5-(4-Chlorobenzylamino)tetrahydropyran-2-yl]-1*H*-imidazole (25) Colorless oil. ^{1}H-NMR (CD_3OD) δ: 1.32—1.58 (1H, m), 1.70—2.09 (2H, m), 2.11—2.29 (1H, m), 2.61—2.79 (1H, m), 3.22—3.33 (1H, m), 4.05—4.17 (1H, m), 4.32—4.41 (1H, m), 6.98 (1H, s), 7.60 (1H, s). {**25**·hydrochloride: $[\alpha]_D$=−28.9° (c=2.6, EtOH)}.

1-Cyclohexyl-3-{(2*S*,5*R*)-2-[1*H*-imidazol-4(5)-yl]tetrahydropyran-5-yl}thiourea (14b) A solution of **1c** (37 mg, 0.22 mmol) and cyclohexyl isothiocyanate (38 mg, 0.27 mmol) in MeOH (6 ml) was refluxed for 3 h. The solvent was evaporated to give a residue that was purified by column chromatography [MeOH–AcOEt (1 : 9)] to yield **14b** (60 mg, 88%) as a colorless oil. $[\alpha]_D$=−9.0° (c=4.6, MeOH). ^{1}H-NMR (CD_3OD) δ: 1.13—2.21 (14H, m), 3.21 (1H, dd, J=10.8, 10.5 Hz), 3.98—4.14 (1H, br), 4.19 (1H, ddd, J=10.8, 5.5, 1.6 Hz), 4.34—4.44 (2H, m), 7.00 (1H, s), 7.61 (1H, s). ^{13}C-NMR (CD_3OD) δ: 26.2, 26.9, 30.9, 31.7, 33.9, 50.7, 53.8, 71.6, 74.4, 116.8, 135.7, 139.1, 181.2. EI-MS m/z: 308 (M^+). HR-MS m/z: 308.1669 (Calcd for $C_{15}H_{24}N_4OS$: 308.1670).

1-(4-Chlorophenyl)-3-{(2*S*,5*R*)-2-[1*H*-imidazol-4(5)-yl]tetrahydropyran-5-yl}urea (15) The same procedure for the preparation of **14b** provided **15** (51%) as an oil. $[\alpha]_D$=−6.8° (c=2.8, MeOH). ^{1}H-NMR (CD_3OD) δ: 1.53—2.22 (4H, m), 3.22—3.38 (1H, m), 3.69—3.88 (1H, m), 4.09—4.20 (1H, m), 4.50—4.64 (1H, m), 7.15—7.40 (4H, s), 7.47 (1H, s). EI-MS m/z: 320 (M^++1). HR-MS m/z: 320.1037 (Calcd for $C_{15}H_{17}ClN_4O_2$: 320.1039).

Animals Male Wistar rats (7—9 week old, Japan SLC, Shizuoka, Japan) weighing approximately 200 g were used. The rats were kept in individual cages on a 12 h light, 12 h dark cycle (lights on at 8:00 to 20:00) and maintained at 25±1 °C with a humidity of 50±10%. Before surgery, they were given free access to standard pellet chow (MF, Oriental Yeast Co., Osaka, Japan) and water. The rats were deprived of food the day before the experiments. All experiments were carried out during the light period in accordance with the Animal Care Committee of the Faculty of Medicine, Osaka University.

***In Vivo* Brain Microdialysis** Rats were anesthetized with urethane (1.2 g/kg, i.p.) and placed in a stereotaxic apparatus (Kopf Instrument, Tujunga, CA, U.S.A.). A microdialysis probe (MAB6; membrane length, 2 mm; ALS/Microbiotech, Stockholm, Sweden) was implanted into the anterior hypothalamic area (AHy)[36] of the rats where histaminergic nerve terminals were most abundant, as illustrated in Fig. 4a, with coordinates of AP, 1.5; L, 0.5; and V, 9.2 mm relative to the bregma, according to the atlas of Paxinos and Watson.[37] AHy was perfused with artificial cerebrospinal fluid (CSF) containing 140 mM NaCl, 3 mM KCl, and 2.5 mM $CaCl_2$, pH 7.4, through the microdialysis probe using a microinfusion pump (CMA100, CMA/Microdialysis AB), and endogenous neuronal histamine in AHy was recovered through the membrane at the probe tip (Fig. 4b). Two hours after insertion of the probe, samples were collected every 20 min with a minifraction collector (CMA140, CMA/Microdialysis AB) and frozen immediately at −40 °C until analysis. The compounds were added to CSF at the concentration of 10 μM and administered through the dialysis membrane. The level of histamine in the perfusate was assayed by an HPLC-fluorometric method[38] (*e.g.*, Fig. 5). In each microdialysis experiment, the average of the first three fractions was defined as basal release, and values of the subsequent fractions were expressed as the percentage of this value. All data are presented as means±S.E.M. in Table 2. Statistically significant differences between basal release and subsequent fractions as well as between groups were analyzed using one-way analysis of variance (ANOVA) with the Newman–Keuls procedure.

Acknowledgments This work was supported in part by Grants-in-Aid for Scientific Research [No. 16590024 (S. H.), No. 16590089 (T. K.)] from the Ministry of Education, Culture, Sports, Science and Technology of Japan.

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