surements depend on the assumption that the dihedral angles of the ethane skeleton are 60° *i.e.*, they refer to rotamers in which the values of θ_2 are either 180 (trans), 60 (gauche), or 300° (gauche). Since the EHT calculations predict that the internal energy of the molecule is minimized at these same values of θ_2, it is permissible to compare directly, at least in an operational manner, the rotamer preferences determined by the two methods.

In conclusion it appears that the EHT-predicted conformer population ratio may reasonably be used to give an indication of the likely situation in aqueous solution for methylhistamines. Its value lies in the possibility for predicting the conformation of molecules, such as side-chain *C*-methyl-substituted histamines, when there are difficulties in deriving the information by experiment.

Acknowledgments. We gratefully acknowledge the contribution made by Drs. G. J. Durant and J. C. Emmett for the synthesis of the methylhistamines and by Dr. P. M. G. Bavin for the synthesis of the piperidines.

References

(1) L. B. Kier, "Molecular Orbital Theory in Drug Research," Academic Press, New York, N. Y., 1971, Chapter VIII.
(2) P. S. Portoghese, *Annu. Rev. Pharmacol*, 10, 51 (1970).
(3) L. B. Kier, *J. Med. Chem.*, 11, 441 (1968).
(4) J-L. Coubeils, Ph. Courrière, and B. Pullman, *C. R. Acad. Sci., Paris, Ser. D.*, 272, 1813 (1971).
(5) S. Margolis, S. Kang, and J. P. Green, *Int. J. Clin. Pharmacol.*, 5 (3), 279 (1971).
(6) P. F. Periti, *Pharmacol. Res. Commun.*, 2, 309 (1970).
(7) A. S. F. Ash and H. O. Schild, *Brit. J. Pharmacol. Chemother.*, 27, 427 (1966).
(8) A. F. Casy, R. R. Ison, and N. S. Ham, *Chem. Commun.*, 1343 (1970).
(9) J. W. Black, W. A. M. Duncan, G. J. Durant, C. R. Ganellin, and M. E. Parsons, *Nature (London)*, 236, 385 (1972).
(10) T. B. Paiva, M. Tominaga, and A. C. M. Paiva, *J. Med. Chem.*, 13, 689 (1970).
(11) J. L. Kavanau, "Structure and Function in Biological Membranes," Vol. 2, Holden-Day, San Francisco, Calif., 1965, p 331.
(12) H. M. Lee and R. G. Jones, *J. Pharmacol. Exp. Ther.*, 95, 71 (1949).
(13) K. Hofmann, "Imidazole and its Derivatives," Interscience, New York, N. Y., 1953, p 188.
(14) B. Pullman, J-L. Coubeils, Ph. Courrière, and J-P. Gervois, *J. Med. Chem.*, 15, 17 (1972), and references cited therein.
(15) (a) R. Hoffman, *J. Chem. Phys.*, 39, 1397 (1963); (b) *ibid.*, 40, 2745 (1964).
(16) J. A. Pople, D. P. Santry, and G. A. Segal, *ibid.*, 43, S129 (1965).
(17) J. A. Pople and G. A. Segal, *ibid.*, 44, 3289 (1966).
(18) M. V. Veidis, G. J. Palenik, R. Schaffrin, and J. Trotter, *J. Chem. Soc. A*, 2659 (1969).
(19) M. J. Adams, D. C. Hodgkin, and U. A. Raeburn, *ibid.*, 2632 (1970).
(20) S. Martinez-Carrera, *Acta Crystallogr.*, 20, 783 (1966).
(21) L. E. Sutton, "Tables of Interatomic Distances and Configurations in Molecules and Ions," *Chem. Soc., Spec. Publ.*, No. 11 (1958); *Suppl.*, No. 18 (1965).
(22) E. L. Eliel, N. L. Allinger, S. J. Angyal, and G. A. Morrison, "Conformational Analysis," Interscience, New York, N. Y., 1965, p 5.
(23) C. Niemann and J. T. Hays, *J. Amer. Chem. Soc.*, 64, 2288 (1942).
(24) S. Castellano and A. A. Bothner-By, *J. Chem. Phys.*, 41, 3863 (1964).
(25) R. J. Abraham and G. Gatti, *J. Chem. Soc. B*, 961 (1969).
(26) J. B. Lambert, R. G. Keske, and D. K. Weary, *J. Amer. Chem. Soc.*, 89, 5921 (1967).

Conformation of Histamine Derivatives. 2. Molecular Orbital Calculations of Preferred Conformations in Relation to Dual Receptor Activity[1]

C. R. Ganellin,*

Smith Kline and French Research Institute, Welwyn Garden City, Hertfordshire, England

G. N. J. Port,† and W. G. Richards

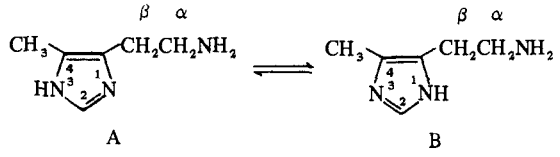
Physical Chemistry Laboratory, Oxford University, South Parks Road, Oxford, England. *Received November 13, 1972*

The EHT calculated trans/gauche conformer population ratios of a series of methylhistamines are compared with their H_1/H_2 histamine receptor agonist activity ratios. Conformer ratios for α- and β-methyl- and *N,N*-dimethylhistamines are predicted to be quite different, respectively 0.1, 0.02, and 4; yet none of these compounds show significant biological selectivity. On the other hand, 2- and 4-methylhistamines have very similar trans/gauche conformational preferences but show a dramatic 1000-fold difference in their activity ratios. Thus, side-chain rotamer preference does not appear to determine whether these conformationally mobile derivatives distinguish between H_1 and H_2 receptors. However, an altered rotamer preference is accompanied by reduced activity and the results are consistent with a trans conformer possibly being involved at both types of histamine receptor.

The physiological actions of histamine can be considered to be mediated by at least two distinct classes of receptor (Ash and Schild).[2] One receptor type, designated H_1 by Ash and Schild, occurs in the smooth muscle of guinea-pig ileum and bronchi and is characterized by the ability of the tertiary amine class of antihistamines to antagonize specifically the response of these tissues. A second receptor type (designated H_2)[3,4] mediates histamine action in stimulating gastric acid secretion, in inhibiting contractions in the rat uterus, and in increasing the heart rate in the guinea pig. The differentiation of these two receptor types is reflected in the relative agonist activities of certain histamine analogs (Black, *et al.*[4]). Thus, 4-methylhistamine‡ selectively stimulates H_2 receptors (the activities, relative to histamine, are in the ratio $H_1:H_2 \approx 1:200$) whereas 2-methylhistamine is signifi-

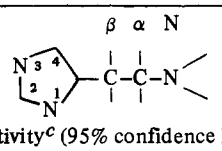
†Supported by the Science Research Council under the CAPS scheme (Cooperative Awards in Pure Science).

‡Using the nomenclature defined in paper 1,[1] 4-methylhistamine represents both tautomeric structure A and B.



A B

Table I. Agonist Activities of Methylhistamines[a]



Compd	Position of Me substituent[b]	Activity[c] (95% confidence limits) relative to histamine = 100		Activity ratio H_1/H_2
		H_1 (ileum)[d]	H_2 (atrium)[e]	
II	2	16.5 (15.1–18.1)	4.4 (4.1–4.8)	4
III	3	0.42 (0.16–1.0)	<0.1[f]	
IV	4	0.23 (0.20–0.27)	43 (40–46)	0.005
V	N	72 (62–84)	74 (71–78)	1
VI	N,N	44 (38–51)	51 (42–61)	1
VIII	β	0.83 (0.42–1.6)	0.89 (0.79–1.1)	1
IX	α	0.36 (0.18–0.73)	0.74 (0.42–1.3)	0.5[g]

[a] Unpublished data provided by Dr. J. W. Black, Head of Pharmacology Group, Smith Kline and French Research Institute. [b] Substituent numbering defined in paper 1. [c] Estimates of relative activity from replicate 2 + 2 parallel line assay. [d] *In vitro*, on guinea-pig ileum, in the presence of atropine. [e] *In vitro*, on guinea-pig right atrium, in the presence of propranolol. [f] Nonparallel dose–response relationship relative to histamine prevents assay. [g] Not significantly different from 1.

cantly less active as a stimulant of H_2 receptors than of H_1 receptors (H_1:$H_2 \approx 4:1$). This extreme sensitivity of activity to an apparently minor structural change in the agonist implies a difference in the chemistry of the receptors and raises the interesting question of whether the chemical involvement of histamine differs at the two types of receptor.

Kier proposed[3] that the dual activity of histamine is a consequence of the ability of its monocation to adopt two distinct and preferred conformations, *viz.* trans and gauche rotamers, and he assigned H_1-receptor activity to the trans rotamer and provisionally associated H_2 activity with the gauche rotamer. As yet, however, there is no corroborative experimental evidence for this proposal. The identification of two different molecular species is suggestive but does not by itself constitute an adequate basis for explaining different biological effects. Although conformation has an obvious importance to drug-receptor interaction, there is no *a priori* reason why conformational effects should be the decisive factor that distinguishes the different receptor effects in the present case. There are other chemical properties of histamine that could be as important.

The postulate that a molecule has to assume a particular conformation in order to function at a receptor implies that the ability of a molecule to achieve the requisite conformation would affect its activity. This is usually interpreted in the sense that activity will be impaired unless a molecule can assume the active form. Kier's identification of minimum energy conformations as being of interest is an important addition to structure–activity considerations because it carries the further implication that a property could be related to a particular population of molecules. The suggestion that a preferred conformation is the active form carries with it the connotation that the concentration of drug required for an effect (*i.e.*, its potency) could be related to the population of drug molecules in the requisite conformation or, for molecules that are conformationally mobile, to the likelihood (*i.e.*, depending on the kinetics of conformer interconversion and drug-receptor interactions) of achieving it.

To relate the biological activity of an agonist to its conformational properties poses considerable problems since it is difficult to devise molecules of a given conformation without also changing some other physicochemical properties. One approach to this problem might be provided by the use of conformationally mobile analogs where the conformational preferences differ from those of histamine. We have considered, as a corollary to the proposal by Kier, that *if minimum energy conformations are of importance, then an altered conformational preference should be accompanied by a corresponding change in biological activity.* Thus, changing the proportion of trans and gauche forms should be reflected by a change in relative activities at the two types of histamine receptor. Such a relationship would also serve to identify which conformation should be associated with action at a particular receptor type.

We have studied the conformational and biological properties of a series of methylhistamines (Table I). Three compounds (II, III, and IV) have the substituent in the nucleus, *viz.* 2-, 3-, and 4-methylhistamines; the others are substituted in the side chain, either on nitrogen (V and VI) or on carbon (VIII and IX). This group of compounds is well suited to consideration of structure–activity relationships. A narrow range of structural modification encompasses a wide range of biological activities of both types H_1 and H_2. The compounds selected all retain conformational flexibility and closely resemble the histamine structure. They would appear to be a homogenous group in regard to those properties, such as molecular weight, partition, and basicity, that are likely to determine access to the active biological site. The methylhistamines have similar pK_a values, lying in the range (at 37°) $pK_{a_1} = 5.8–6.8$, $pK_{a_2} = 9.1–9.8$ § and the introduction of a methyl group is not expected to have a marked effect on the tautomeric properties. Compound III is an exception in being unable to undergo tautomerism but it is structurally analogous to the tautomeric form on which the present considerations are based.

Biological Activities. The H_1 and H_2 activities were assessed under comparable circumstances by being estimated *in vitro*, on isolated tissues obtained from the same animal species, to minimize the complicating effects of drug transport and metabolism. This is important since our discussion depends on the assumption that differences in the activities of the compounds are due to differences in drug-receptor interactions rather than to differences in access. For each compound the agonist activity relative to histamine was determined on both guinea-pig ileum (H_1) and guinea-pig atrium (H_2). The activities in Table I are expressed on a molar basis relative to histamine, rather than as absolute concentrations, and should therefore be unaffected by differences between the sensitivity of the tissues in their response to stimulation.

Although methyl substitution reduced activity the compounds qualitatively resembled histamine and afforded

§ Unpublished data, Smith Kline and French Research Institute.

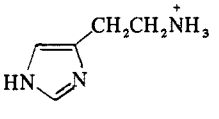
Table II. Energies by EHT of Minimum Energy Conformations for Methylhistamine Monocations

	Trans (θ_2 = 180°)		Gauche (θ_2 = 300°)				
Molecule	θ_1, deg	Total energy, eV	θ_1, deg	ΔE^b	θ_1, deg	ΔE^b	$n_t{}^a$
Histamine	120	−799.600[h]	120	0.3	270	1.0	0.55
II	120	−903.954[h]	120	0.3	270	1.0	0.55
III	120	−904.297[h]	120	0.3	270	1.0	0.55
IV	180	−903.722[c,h]	150	1.0[c]	270	1.2[c]	0.75
V	120	−904.268[h]	120	0.3	270	1.0	0.55
VI	120	−1008.106[h]	120	0.9	300	1.8	0.78
VIII	120	−903.748[d]	120	−2.3[d,h]	300	−0.3[e]	0.02
IX	120	−903.964[f]	120	−1.2[g,h]	270	−0.6[g]	0.09

[a]Population of trans conformer (mole fraction) at 37° [b]Energy relative to trans conformation (the most stable for compounds I–VI) in kilocalories per mole. [c]θ_4 = 120° [d]$\theta_1{}^{Me}$ = 240°. [e]$\theta_1{}^{Me}$ = 60°. [f]$\theta_2{}^{Me}$ = 300°. [g]$\theta_2{}^{Me}$ = 180°. [h]Most stable conformation for each compound.

linear dose–response relationships which paralleled that of histamine, *i.e.*, activity was linearly concentration dependent. Compound III (3-methylhistamine) was an exception in giving a nonparallel (flatter) dose–response relationship relative to histamine at the H_2 receptor. The implication is that it possesses a lower intrinsic activity than does histamine and for this reason we have not assigned a figure to its activity ratio. As indicated in Table I there is no significant difference (within 95% fiducial limits) between the compounds V–IX in their relative activities at the two receptors, *i.e.*, $H_1/H_2 = 1$. However, it is not inherent in the biological system that the two types of receptor necessarily respond equally well to stimulation by histamine derivatives, as demonstrated by compounds II and IV. These do show differences which are highly significant. 4-Methylhistamine (IV) is much less active than 2-methylhistamine (II) at the H_1 receptor, as has been noted previously,[5] and a new point of interest is that the converse holds at the H_2 receptor.

Conformational Analysis and Theoretical Predictions. The conformational preferences for the monocations are derived from the molecular energies of the most stable conformations as determined by extended Hückel theory (EHT) calculations, it having been established[1] that EHT calculations provide a predictive model for conformer population ratios of methylhistamines in aqueous solution. The compounds form a homogenous set for calculation since the same parameters can be used throughout. There is a special advantage in studying a series of similar molecules; it permits an analysis of a chemical property relative to the biological reference material, histamine, and thereby avoids some of the problems associated with assessing the validity of absolute data derived from a single species. Since the EHT predictions for histamine did not differ in substance between the dication or either tautomer of the monocation,[1] we have concentrated on the N_3–H tautomeric form of the monocation (I), the form considered by Kier and the most prevalent[#] of the different species, for the present study.



I

The molecular energies of the most stable conformations, and relative energies of the two conformations next in stability, as determined by EHT calculations for each of the monocations are listed in Table II. For 2-, 3-, and *N*-methylhistamine monocations (II, III, and V) the methyl group is found to have no appreciable effect on the conformation

[#]It should, however, be noted that for $pK_{a_1} > 6.0$ (compounds II and IV) the dication predominates at pH 6.0.

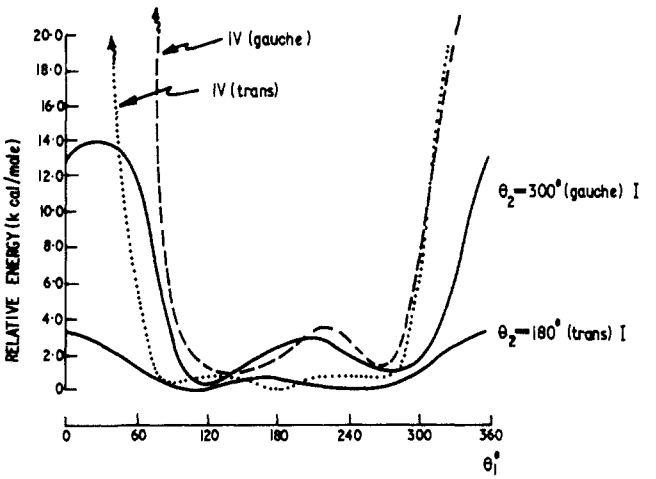


Figure 1. Histamine (I) and 4-methylhistamine (IV) monocations. Relative energies (from EHT) of trans and gauche rotamers showing the influence of ring orientation θ_1 The orientation θ_4 of the methyl group in IV is 120°.

relative to histamine. With 4-methylhistamine monocation (IV), the stable conformations are predicted to be staggered (*viz.* when θ_2 = 180 or 300°) and the values of θ_1 for minimum energy to be 180° (when θ_2 = 180°) or 150 and 270° (when θ_2 = 300°). Of the two likely orientations, θ_4 = 60 or 120°, for the methyl group it is more favorable in all cases to have θ_4 = 120°. The trans conformation is predicted to be the more stable, by about 1 kcal/mol, suggesting that it would be slightly more preferred than with histamine, populating the trans rotamer to the extent of about 75%. It should, however, be noted that the nmr results derived from aqueous solution suggest[1] that there is no appreciable difference in the rotamer populations between histamine monocation and its 4-methyl derivative. The essential conclusion remains that the methyl group causes no real disturbance of rotamer population even though it has a marked influence on rotational barriers and alters considerably the values permitted for θ_1 Profiles showing the influence of ring orientation on the rotamer energies of 4-methylhistamine are compared with the analogous profiles for histamine monocation in Figure 1; the methyl group raises the rotational barriers in the region $60° > \theta_1 > 300°$, presumably as a consequence of interaction between the methyl group and the α-methylenic protons. Another difference between the profiles probably derives from interactions which would occur, but to a lesser extent, between the methyl group and the β-methylenic protons; it appears that for the trans form (θ_2 = 180°) of 4-methylhistamine monocation, energies in the region of θ_1 = 120° (an energy minimum for the dication or for histamine mono- and dicat-

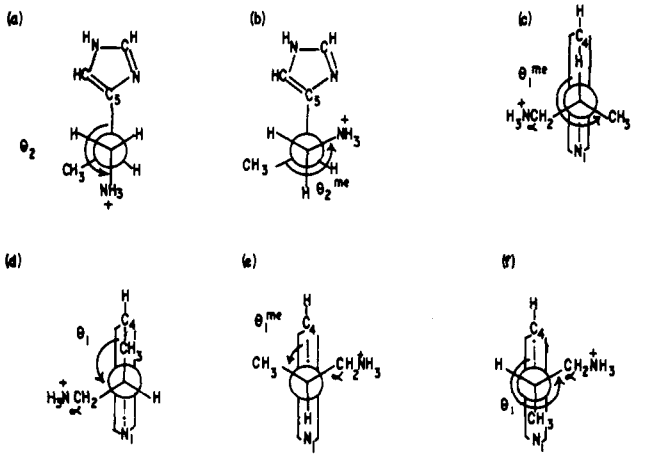


Figure 2. β-Methylhistamine (VIII) monocation showing (i) torsion angles θ_2 and θ_2^{Me} for the most stable conformations viewed along C_α–C_β bond from C_α toward C_β (a) trans, $\theta_2 = 180°$, $\theta_2^{Me} = 60°$; (b) gauche, $\theta_2 = 300°$, $\theta_2^{Me} = 180°$; and (ii) torsion angles θ_1 and θ_1^{Me} and view along C_β–C_5 bond, looking from C_β to C_5 with (c) $\theta_1 = 120°$, $\theta_1^{Me} = 240°$; (d) $\theta_1 = 120°$, $\theta_1^{Me} = 0°$; (e) $\theta_1 = 300°$, $\theta_1^{Me} = 60°$; (f) $\theta_1 = 300°$, $\theta_1^{Me} = 180°$.

ions) are relatively higher, with the net result that the minimum shifts to $\theta_1 = 180°$, the form in which all the carbon and nitrogen atoms are coplanar. For *N,N*-dimethylhistamine (VI), the energies of the two stable gauche forms are predicted to be around 1–2 kcal/mol greater than for the trans, implying that about 80% of molecules would be in the trans form.

The two remaining side-chain substituted compounds have a *C*-methyl group. Each can be analyzed with reference to histamine and, provided that it can be assumed that the methyl group adopts a staggered configuration with respect to the substituted carbon atom, the problem reduces to the minimizing of energy as a function of the two angles θ_1 and θ_2. With β-methylhistamine (VIII), shown in Figure 2, the main difference from histamine arises through interaction between the methyl and ammonium groups; the relative energies for rotamers in which the ammonium group is either (a) trans to imidazole ($\theta_2 = 180°$) or (b) trans to methyl ($\theta_2 = 300°$) are calculated for different values of θ_1, remembering that there are now two sets of conformers for each value of θ_1 because of the asymmetry introduced at the β-carbon atom. The most stable conformation is predicted to be the gauche** form, $\theta_1 = 120°$, $\theta_2 = 300°$, where the methyl group has $\theta_1^{Me} = 240°$ (Figure 2c) rather than 0° (Figure 2d). This conformation has an energy at least 2 kcal/mol lower than any of the other stable forms. The most stable trans rotamer similarly has $\theta_1 = 120°$, $\theta_2 = 180°$, and $\theta_1^{Me} = 240°$ and is predicted to be populated perhaps to the extent of about 2% of the molecules. A second stable gauche form, also energetically more favorable than the trans, has $\theta_1 = 300°$, $\theta_2 = 300°$, where $\theta_1^{Me} = 60°$ (Figure 2e) rather than 180° (Figure 2f). For α-methylhistamine (IX) differences from histamine arise mainly as a consequence of interactions between the ring and methyl substituent; the EHT calculations predict that it is less favorable to have the ammonium group than the methyl group trans to the ring, to the extent that the trans rotamers are less preferred. The most stable conformation predicted has $\theta_1 = 120°$, $\theta_2 = 300°$, and $\theta_2^{Me} = 180°$ (see Figure 3 for θ_2^{Me}). The energetically nearest other

**Unless otherwise stated, trans and gauche, in the ensuing discussion and in the tables, denote the configuration of the ammonium group relative to imidazole.

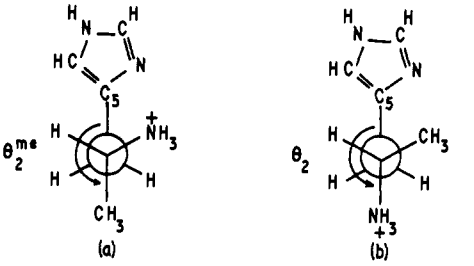


Figure 3. α-Methylhistamine (IX) monocation showing torsion angles θ_2 and θ_2^{Me} for most stable conformations viewed along C_α–C_β bond from C_α toward C_β: (a) gauche, $\theta_2 = 300°$, $\theta_2^{Me} = 180°$; (b) trans, $\theta_2 = 180°$, $\theta_2^{Me} = 300°$.

Table III. Comparison of Trans/Gauche Conformer Ratio with H_1/H_2 Activity Ratio for Methylhistamines

Molecule[a]	Most stable conformation,[b] θ_1/θ_2, deg	Conformer ratio, trans/gauche[c]	Activity ratio,[a] H_1/H_2
Histamine	120/180 trans	1.2	1
II	120/180 trans	1.2	4
III	120/180 trans	1.2	
IV	180/180 trans	3	0.005
V	120/180 trans	1.2	1
VI	120/180 trans	3.5	1
VIII	120/300 gauche	0.02	1
IX	120/300 gauche	0.1	1

[a]Position of methyl substituent and activity ratio from Table I. [b]Energies given in Table II. [c]Derived from n_t of Table II.

stable gauche conformer has $\theta_1 = 270°$, $\theta_2 = 300°$, and $\theta_2^{Me} = 180°$. The most stable trans rotamer has $\theta_1 = 120°$, $\theta_2 = 180°$, and $\theta_2^{Me} = 300°$ and is less preferred than either of the gauche forms. Energy differences for these conformers, as given in Table II, suggest that the population of molecules in a trans form would be about 10%.

Discussion

The influence of a nuclear methyl substituent on conformer preference is small and interest centers particularly on the effect of side-chain methylation, where a *C*-methyl group is predicted to lead to substantial energy differences between the trans and gauche forms and, indeed, to a reversal of the relative stabilities. These compounds therefore provide an exeellent opportunity to examine conformation–activity relationships and in particular to seek evidence for that proposed by Kier. If different conformations are required at the two receptor types, and if activity depends on the concentration of the relevant conformer, then the ratio of H_1 and H_2 activities should parallel the population ratio of the two conformations concerned. Thus, positive evidence for Kier's postulate would be provided if α- and β-methylhistamines, being largely in gauche conformations, were found to be relatively more active at the H_2 receptors, and *N,N*-dimethylhistamine (VI), which relative to histamine has a little more preference for being in a trans form, showed some disposition toward activity at H_1 receptors.

The predicted population ratio of trans/gauche conformers is compared in Table III with the ratio H_1/H_2 of activities found at the two receptors. They appear to be unrelated. The side-chain substituted compounds (VI, VIII, and IX) predicted to be conformationally different from histamine show no selectivity of action. Even though the introduction of a side-chain *C*-methyl group considerably reduces the potency relative to histamine, it does so to an almost equal degree at the two receptors. Conversely, the ring-substituted

compounds (II and IV) which do show a dramatic selectivity of action still have a rotamer composition similar to that of histamine. Many factors may affect activity in this series of compounds. These results suggest that trans/gauche rotamer preference, as predicted by EHT calculation, does not appear to be of major importance in determining whether these conformationally flexible molecules distinguish between H_1 and H_2 histamine receptors.

Since the side-chain substituted compounds differ in conformational preference, one can examine whether they show a consistent relationship between activity and rotamer preference at either of the histamine receptors. In fact, a qualitative pattern is apparent since the two *N*-methyl compounds (V and VI) which resemble histamine in rotamer preference are nearly as active as histamine, whereas the two *C*-methyl compounds (VIII and IX) for which the trans rotamer is no longer preferred are much weaker in activity. Their reduced activity would be due to a variety of causes, one of which could be a reduced preference for the trans conformation. Insofar as there is a self-consistency in these observations it suggests that a trans conformer of the agonist may be associated with activity at both types, H_1 and H_2, of histamine receptor. This does not prove that the trans conformer is required and it would be premature to conclude that the gauche form of histamine is not involved in receptor interaction.

Acknowledgments. We gratefully acknowledge our debt to Dr. J. W. Black for his guidance during many stimulating discussions and thank him for permission to quote results.

References

(1) C. R. Ganellin, E. S. Pepper, G. N. J. Port, and W. G. Richards, *J. Med. Chem.*, 16, 610 (1973) (paper 1).
(2) A. S. F. Ash and H. O. Schild, *Brit. J. Pharmacol. Chemother.*, 27, 427 (1966).
(3) L. B. Kier, *J. Med. Chem.*, 11, 441 (1968).
(4) J. W. Black, W. A. M. Duncan, G. J. Durant, C. R. Ganellin, and M. E. Parsons, *Nature (London)*, 236, 385 (1972).
(5) R. G. Jones in "Handbook of Experimental Pharmacology," Vol. XVIII/I, O. Eichler and A. Farah, Ed., Springer-Verlag, Berlin, 1966, Chapter 1.

Conformation of Histamine Derivatives. 3. A Relationship between Conformation and Pharmacological Activity

C. R. Ganellin

Smith Kline and French Research Institute, Welwyn Garden City, Hertfordshire, England. *Received November 13, 1972*

The methyl substituent of 4-methylhistamine affects the conformation of the molecule by influencing the orientation of the imidazole ring with respect to the side chain and introducing a measure of rigidity through restricting rotation. Implications are discussed regarding biological activity at the H_1 and H_2 histamine receptors. The dramatic receptor selectivity of 4-methylhistamine is accounted for in the difference between its conformational properties and those of histamine. It is proposed that for histamine a conformation essential to productive interaction with the H_1 receptor is one in which the side chain is fully extended (trans form) and all carbon and nitrogen atoms are coplanar with the imidazole ring. In this conformation the separation between the ring N_1 and ammonium nitrogen atoms is at the maximum distance of 5.1 Å.

The marked effect on biological activity caused by methylation at different positions in the histamine molecule[1,2] (compounds II–IX in Table I) poses an intriguing medicinal chemical problem. Depending on its position, the presence of the methyl group can result in as much as 1000-fold reduction in histamine-like stimulant activity or be almost without influence. It can also lead to a selectivity such that a molecule appears to be less active at one type of histamine receptor than at another, as, for example, with 2-methylhistamine (II) which is significantly less active as a stimulant of H_2 receptors than of H_1 receptors (the activities, relative to histamine, are in the ratio H_1:$H_2 \approx 4:1$) and 4-methylhistamine (IV) which, conversely, is considerably less active at H_1 than at H_2 receptors (H_1:$H_2 \approx 1:200$).[2] Indeed, the very dramatic influence of the substituent in 4-methylhistamine makes this molecule of especial interest. There is every reason to suppose that for these compounds the differences in activities are on the whole due to differences in drug-receptor interactions,[1,2] so that one may reasonably argue that the presence of the methyl substituent somehow affects the chemistry of the interaction process. In the absence of any molecular description of the receptor one is left to examine the drug molecule, initially to define the chemical consequence of changing the drug structure and subsequently to establish which changes in chemical properties affect biological activity. It could be argued that the methyl substituent would either affect the chemical reactivity of the drug molecule or act sterically to hinder drug-receptor interaction, but then one should adduce supporting evidence and also identify more specifically the manner in which this could occur. To this end we have examined whether the methyl group might alter, *inter alia*, the conformation of the drug molecule.

In a previous paper[1] we reported our findings and considered whether there was evidence among a series of conformationally mobile methylhistamines that the side-chain conformation might determine their ability to stimulate H_1 and H_2 receptors. There was no indication from our results that alteration of rotamer preference affected selectivity but there was a suggestion that it might lead to reduced activity. Conversely, the compounds that showed selectivity did not differ from histamine in their side-chain rotamer preference. It therefore appears that properties other than rotamer preference in the side chain distinguish activity at the two receptor types. The possibility remains that activity is influenced by other conformational effects. In the present paper we examine whether the pronounced selectivity of 4-methylhistamine could be due to a conformational influence of the methyl substituent and, as a corollary, whether one may therefore infer an active conformation for histamine. It must be noted that *we cannot by this means assert that conformation definitely determines biological activity, but we can indicate whether the observed changes in activity accompany-*