Conformational Analysis of Cyproheptadine Hydrochloride

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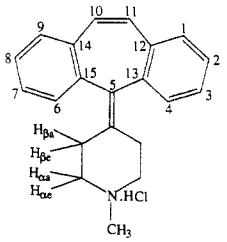
A nuclear magnetic resonance and theoretical study on the conformations and molecular flexibility of cyproheptadine hydrochloride (1) is reported. In the ^1H NMR spectrum of 1 in $CDCl_3$, two conformational forms are observed to occur in an approximate ratio of 1:4. In both forms, NOE and coupling constant measurements suggested that the terminal *N*-methyl group is equatorial. NOE experiments identified the more populated conformer (labeled D) as similar to the form seen in the X-ray crystal structure of cyproheptadine. The other form observed (A) may in principle be converted to D via either inversion of the central ring (T_{inv}) or concerted nitrogen (N_{inv}) and piperidine ring inversion (P_{inv}). Chemical-exchange peaks in the 400-MHz 2D NOESY/chemical-exchange spectrum suggested that the latter mechanism is responsible for interconversion between the two forms. A theoretical study of the various interconversion processes using both molecular mechanics (MM2) and molecular orbital (AM1) approaches is also reported.

I. Introduction

Cyproheptadine hydrochloride (1), a drug currently marketed as an antihistaminergic, is also used with moderate success in the symptomatic treatment of a number of other disorders such as Cushing's disease, Parkinsonism, and Nelson's syndrome and other carcinoma-related syndromes.[1] Such a wide variety of applications can be attributed to the fact that cyproheptadine is almost equipotent[2] at three major neuroreceptors in both the peripheral[3,4] and central[2] nervous systems: acetylcholinergic (muscarinic M-1[5]), histaminergic (H-1[6]), and serotonergic (5HT-2[7,8]). It also enhances appetite and so is used for promoting weight gain through increased food consumption.[1]



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Cyproheptadine (1) contains a tricyclic nucleus with a 1-methyl-4-piperidylene ring attached to the central heptatriene ring. It is a relatively simple structure, devoid of characteristic binding groups (apart from the aromatic ring and the nitrogen) capable of specific interactions with the receptor, which probably accounts for its nonselectivity. Its shape is similar to that of a number of antidepressant drugs (e.g., imipramine, mianserin), with which it shares some common pharmacological properties such as binding to central nervous system (CNS) 5HT-2 receptors.[2,7]

Because of its structural simplicity and relative rigidity, cyproheptadine is a suitable structural nucleus on which to attach additional functional groups in order to modify its pharmacological profile. The synthesis and testing of various cyproheptadine derivatives have shown that such an approach may lead to the discovery of highly potent and selective compounds; for example, (i) saturation of the ethylene bridge and 3-COOH substitution leads to a significant reduction in antiserotonergic and anticholinergic properties while its potency at the histamine receptor and its appetite-stimulating properties[9] are retained. (ii) Saturation of the ethylene bridge and replacement of one of the methylenes by an oxygen (as in Danitracen[1]) results in loss of anticholinergic activity and enhancement of its antidepressant and sedative effects. (iii) In L-646,462,[10] which has a number of substitutions, antiserotonergic activity is halved compared with that of cyproheptadine, indicating a new role as a long-lasting, peripherally selective dopamine antagonist. (iv) Various substitutions at the 3-position of the tricyclic ring lead to enhancement of dopaminergic and α-adrenergic blocking properties, while anticholinergic activity is only partially retained. This substitution also results in differentiation of the activities of the resulting enantiomers of these 3-substituted derivatives.[11-13]

For more effective drug design based on cyproheptadine, a knowledge of its conformational flexibility and the geometries of its stable conformers is essential. Apart from the introduction of new potential binding groups, substitution can also modify the geometry of the basic ring system by introduction of an additional strain into the rings. The consequences of such changes may be an in-

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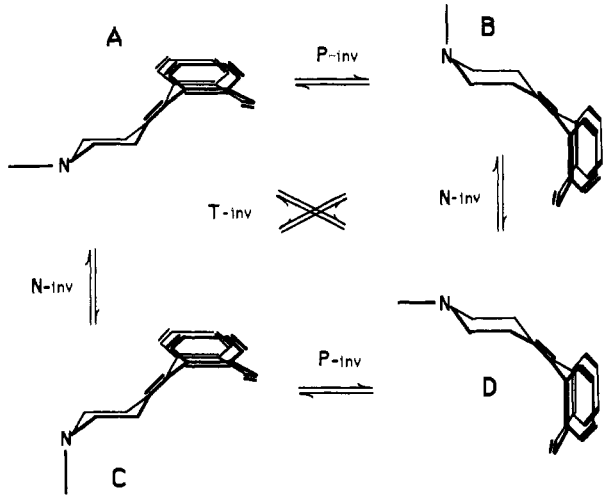

Figure 1. Three major conformational processes between stable conformers of cyproheptadine.

crease or decrease in the interconversion barriers between the possible conformers, stabilization or destabilization of a particular conformer, and modification of the pK_a of the nitrogen in the piperidine ring.

The structure of cyproheptadine, determined by X-ray crystallography,[14] has been used to extract relevant geometrical parameters (e.g., the distance of nitrogen from the centers of the aromatic rings or from the planes of those rings) for comparison of the structures and bioactivity of cyproheptadine and other tricyclic compounds.[5,15] This structure has also been used to model the pharmacophore of H-1 receptor antagonists.[15,16]

It is generally assumed[5,15] that cyproheptadine is a rigid compound with its bioactive conformation probably close to its X-ray structure. This implies that the three receptors H-1, M-1, and 5HT-2 have a common portion of the binding site complementary to this cyproheptadine conformation. Alternatively, it can be argued that cyproheptadine in solution may exist in different conformations, each responsible for one or more of its characteristic bioactivities. The latter possibility prompted us to investigate the behavior of cyproheptadine in solution using nuclear magnetic resonance (NMR) techniques, assisted by some theoretical calculations and computer graphics.

Conformational Possibilities for Cyproheptadine. Having a pK_a of 9,[8] the nitrogen atom in the piperidine ring of cyproheptadine is protonated at physiological pH. The rate of proton exchange will therefore influence barriers to the associated conformational processes available: piperidine ring inversion (P_{inv}) and nitrogen inversion (N_{inv}). A third conformational process, tricyclic ring inversion (T_{inv}), is expected to be relatively free from the influence of proton exchange. These three processes can (considering only the chair conformation for the piperidine ring and the boat conformation for the central heptatriene ring as stable conformations) give rise to eight conformations. Because of the molecular symmetry along the mirror plane passing through the N1 and C4 atoms of the piperidine ring, these reduce to four unique conformations, A, B, C and D (Figure 1) containing only two distinct conformational forms, A,C and B,D of the tricyclic/piperidine ring system. The X-ray-determined structure[14] corresponds to conformation D.

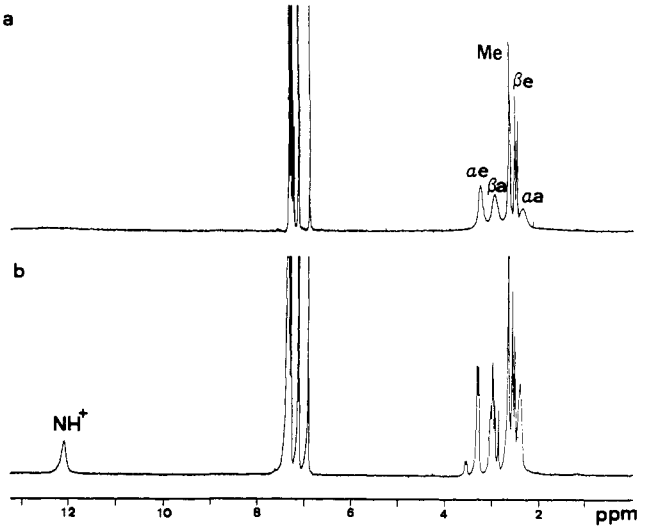

Figure 2. (a) 300-MHz ^1H NMR spectrum of cyproheptadine hydrochloride in ultrapure $CDCl_3$ at room temperature. (b) Spectrum of the same sample at 230 K.

Any substitution in the cyproheptadine rings destroys the molecular symmetry and extends the possible number of conformers because of introduced chirality. This type of chirality, derived from molecular asymmetry rather than the presence of asymmetric centers, is called atropisomerism.[17] The separation of chiral isomers of cyproheptadine derivatives and their independent evaluation for bioactivity[11-13] is important in view of the commonly observed stereochemical dependence of receptor binding.[18]

II. Conformational Study Using NMR

Experimental Methods. The NMR experiments were performed for solutions (~30 mg/mL) of cyproheptadine hydrochloride in $CDCl_3$, CD_2Cl_2 and MeOD-d_3. The digital resolution for ^1H chemical shifts (referenced to TMS) was 0.01 ppm, and for ^{13}C chemical shifts (referenced to the center of the $CDCl_3$ triplet, 77.0 ppm from TMS, or the center of the MeOD-d_3 multiplet, 49.0 ppm from TMS), it was 0.05 ppm. Coupling constants are reported to ±0.5 Hz. 1D standard spectra, selective decoupling, NOE, and variable-temperature experiments were done mainly on a Bruker AM-300 spectrometer at 300.12 MHz for the ^1H experiments and 75.4 MHz for the ^{13}C experiments. Some additional 1D experiments and the 2D NOESY/chemical-exchange experiment were done on a Varian VXR-400 spectrometer at 399.9 MHz. The 2D NOESY experiment was performed in phase-sensitive mode using the hypercomplex method,[19] with a 400 ms mixing time, a spectral width of 3000 Hz with quadrature detection in both dimensions, 1024 points in t_2 for each of the 512 t_1 values, and a relaxation delay of 1.5 s. The data were zero-filled to 1024 points in t_1 prior to Fourier transformation. To avoid truncation of free induction decays, a squared sine-bell apodization function was used to weight the data.

Results and Discussion. To reduce proton exchange in solution, relatively nonpolar solvents, namely chloroform

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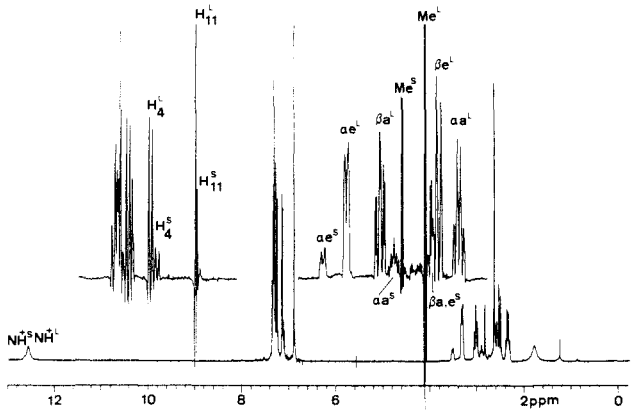

Figure 3. 400-MHz ^1H NMR spectrum of cyproheptadine hydrochloride in $CDCl_3$ (with a trace of acid present) at room temperature with insets of expanded (and resolution-enhanced) aromatic and aliphatic regions.

and methylene dichloride, were chosen for the initial NMR studies. In these solvents the ^1H NMR spectrum of cyproheptadine hydrochloride at room temperature was sometimes characterized by broad lines in the aliphatic region (Figure 2), indicative of the presence of some exchange process, and sometimes by sharp multiplets in the same region. This anomaly was later attributed to the acidity and water content of the solvent used, broad lines being observed in ultrapure deuterated solvents and sharp lines occurring in solvents which may have contained traces of acid or in deliberately HCl-acidified solutions. Recovered samples of cyproheptadine from each type of solution were shown by mass spectroscopy to be identical with the original cyproheptadine hydrochloride.

The spectrum in Figure 2a is characteristic of a sample run in ultrapure deuterated solvent (treated with dry silica gel) containing no traces of acid or water. It consists of three broad humps in the aliphatic region (3.27, 2.98, 2.39 ppm), with a sharp singlet (2.65 ppm) and a sharp doublet (2.52 ppm) overlapped with the highest field hump. Integration of this region gives a ratio of 2:2:7. In the aromatic region there is a sharp singlet for the ethylene bridge at 6.88 ppm, a doublet at 7.10 ppm, and a second-order splitting pattern at 7.2–7.5 ppm, corresponding to the remaining aromatic protons (integration 2:2:6). No resonance for the NH^+ proton is observed in the range 0–15 ppm. The presence of chemical exchange was confirmed by cooling the sample to 230 K. At this temperature the broad humps are resolved into a broad doublet, a triplet, and a quartet, corresponding to $H_{\alpha e}$ (geminal coupling), $H_{\beta a}$ (geminal and axial–axial coupling), and $H_{\alpha a}$ (geminal, axial–axial, and axial–NH_{ax} coupling), respectively. The sharp singlet and doublet noted previously are assigned to the N–Me and $H_{\beta e}$ (geminal coupling) protons, respectively. The NH^+ resonance was also observed at this temperature at 12.1 ppm (Figure 2b). This assignment was confirmed by selective decoupling at low temperature. Also of interest is the emergence of additional peaks which include a doublet at 3.55 ppm, a singlet at 2.85 ppm, and a broad hump on the low-field side of the NH^+ resonance (Figure 2b), which at this stage were suspected of belonging to some other conformer of cyproheptadine. This was confirmed from spectra recorded under acidic conditions (discussed later, Figure 3), which show fine splitting at room temperature.

The doublet at 7.10 ppm in the aromatic region was identified as belonging to H_4 (vicinal to the exocyclic double bond) because irradiation of the bridge protons prior to acquisition leads to an NOE[20] enhancement of a

Table I. ^1H Chemical Shifts[a] (ppm, Referenced to TMS) and Coupling Constants (Hz)[b] for Hydrogens on the Piperidine Ring in Cyproheptadine Hydrochloride in Acidified $CDCl_3$

hydrogen	^1H chemical shift L	^1H chemical shift S	multiplicity[c]	$^2J_{gem}$	$^3J_{aa}$	$^3J_{ee}$	$^3J_{ae}$	$^3J_{NH-H_\alpha}$
$H_{\alpha a}$	2.35	2.90	quartet	11.5	13.0		3.5	9.5
$H_{\alpha e}$	3.31	3.51	doublet	11.5		4.5	5.0	2.5
δ_{ae}[d]	0.96	0.61						
$H_{\beta a}$	3.05	2.55	triplet[e]	15.0	13.0		5.0	
$H_{\beta e}$	2.52	2.55	doublet[e]	15.0		4.5	3.5	
δ_{ae}[d]	0.53							

[a] Chemical shifts are given for L and S conformers. [b] Coupling constants are for the L conformer, expressed to ±0.5 Hz. [c] Main overall appearance, not including fine coupling. [d] δ_{ae} values are shift differences between axial and equatorial hydrogens. [e] Overlap of these resonances in the S conformer results in an apparent multiplet.

doublet at 7.28 ppm, hidden among the overlapped aromatic proton resonances. This doublet was assigned to the H_1 proton because of its proximity to the ethylene bridge. By a combination of selective decoupling, NOE experiments, and resolution enhancement of the spectra, assignments in the aromatic region were made (H_1, 7.28 ppm doublet, $^3J_{ortho}$ = 7.5 Hz; H_2, 7.22 ppm triplet, $^3J_{ortho}$ = 7.5 Hz, $^4J_{meta}$ = 1.5 Hz; H_3, 7.29 ppm triplet, $^3J_{ortho}$ = 7.3 Hz, $^4J_{meta}$ = 1.8 Hz; H_4, 7.1 ppm doublet, $^3J_{ortho}$ = 7.2 Hz). These were confirmed by simulation of this section of the spectrum using the Bruker utility program PANIC.

On return to room temperature, the aliphatic region again attained the characteristic broad lines. The broad-to-sharp-to-broad change was fully reversible and repeatable with the same sample (this was checked over a period of 6 weeks). Heating of the sample above room temperature to 330 K did not lead to any other line-shape changes, although some shifts of peaks in the aromatic region (H_4 and H_{11}) were observed. Variable-temperature experiments indicated a coalescence temperature of about 260 K for the singlets at 2.65 and 2.85 ppm assigned to the NMe groups of the two conformers. The rate of interconversion at this temperature was estimated from eq 1 and used in the Eyring equation[21] to calculate an inversion barrier (whatever the process) of about 12.6 kcal/mol.[22]

$$k = \frac{\pi \delta \nu}{\sqrt{2}} \quad (1)$$

The pattern observed in the aliphatic region (Figure 2b) at low temperature is obtainable at room temperature if the sample is slightly acidified. The exact chemical shifts and proportion of conformers are, however, dependent on pH, water content, and temperature. The 400-MHz spectrum of cyproheptadine chloride at room temperature in acid-treated $CDCl_3$ (Figure 3) clearly shows the presence of the other conformer, including two broad lines for NH^+ (12.58 and 12.69 ppm), two doublets for H_4 (7.12 and 7.10 ppm, $^3J_{ortho}$ = 7.5 Hz) two sharp lines for the bridge protons (6.89 and 6.88 ppm) and two multiplets for each of the protons in the aliphatic region. At this resolution the two

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(22) Note that eq 1 applies strictly only for the case of equally populated two-site exchange. In the present case the population ratio is approximately 4:1 and use of a modified treatment yields energy barriers within 1 kcal/mol of the value obtained with the two-site expression in eq 1. (Shanan-Atidi, H.; Bar-Eli, K. H. *J. Phys. Chem.* **1970**, *74*, 961.)

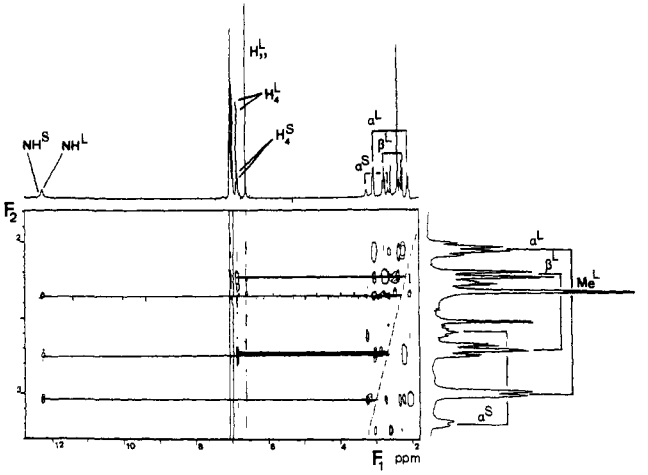

Figure 4. Upper section of phase-sensitive 2D NOESY spectrum (400 MHz) of cyproheptadine hydrochloride under acidic conditions at room temperature. In the phase-sensitive mode shown, the diagonal has a negative intensity and NOE cross-peaks are positive. Only positive contours are shown on the plot. A dashed line indicates the diagonal; solid lines indicate the NOE cross-peaks (from the top H_4^L–$H_{\beta e}{}^L$, NH^L–Me^L, $H_4{}^L$–$H_{\beta a}{}^L$, NH^L–$H_{\beta a}{}^L$, and NH^L–$H_{\alpha e}{}^L$) which identify the more abundant conformer, L.

NMe resonances appear as doublets (^3J = 4.5 Hz), split by coupling to the NH^+ protons, which exchange very slowly under acidic conditions. Selective saturation of the NH^+ peak in each conformer leads to the collapse of the corresponding NMe doublets (2.86 and 2.61 ppm) to singlets, and to the collapse of apparent quartets for the $H_{\alpha a}$ protons (2.90 and 2.35 ppm, $^3J_{NH-H}$ = 9.5 Hz) to triplets. A large coupling observed for NH^+–$H_{\alpha a}$ in both conformers indicates that the NMe group is in an equatorial position in each conformer. Selective decoupling and resolution enhancement lead to the remaining assignments in Table I. In both conformers the couplings within the piperidine ring are consistent with a chair conformation of this ring.

The ratio of the two conformers, designated S (small) and L (large) in the following discussion, is approximately 1:4. These S and L conformers must be associated with structures A and D. Two questions are of interest: which conformer is A and which is D, and by what conformational process are they related?

Computer-graphic examination of A and D indicates that NOE's between the aromatic and aliphatic protons might be used to identify the S and L conformers. Structure A has the H_4 proton (a well-resolved doublet in the aromatic region) within 4 Å of $H_{\alpha a}$ and $H_{\beta e}$ whereas in structure D only $H_{\beta e}$ and $H_{\beta a}$ are within this distance of H_4. The 2D NOESY spectrum (a section of which is shown in Figure 4) shows cross-peaks between H_4 and both β-protons but not to any α-protons, thus indicating that it is the more abundant conformer that corresponds to structure D (i.e., that found by X-ray crystallography[14]).

Additional evidence for this assignment comes from a striking difference between the $H_{\alpha a}$ and $H_{\alpha e}$ proton chemical shifts (δ_{ae}) in these two conformers which is not reflected in the H_β shifts. While δ_{ae} for the S conformer (0.61 ppm) is similar to that of other protonated NMe piperidines,[23] δ_{ae} for the L conformer (0.96 ppm) corresponds to the value usually quoted for unprotonated NMe equatorial piperidines (the larger value in the unprotonated compounds being explained by the additional shielding of $H_{\alpha a}$ by the lone pair on the nitrogen in the axial position). Examination of structures A and D by computer graphics shows that only the $H_{\alpha a}$ protons in the D conformer are within the shielding influence of the ethylene bridge, which can cause an extra high-field shift[24] for these protons in cyproheptadine hydrochloride. Thus the L conformer is likely to be structure D.

Table II. ^{13}C Chemical Shifts (ppm, Referenced to $CDCl_3$ at 77.0 ppm from TMS) of Cyproheptadine Hydrochloride in Acidified $CDCl_3$ at Room Temperature (L and S Conformers) and in Pure $CDCl_3$[a]

carbon	chemical shift			
	L conformer	S conformer	pure $CDCl_3$	MeOD-d_3
C_1	128.32*	128.60	128.36*	129.58
C_2	126.99	127.12	126.95	128.15
C_3	128.25*		128.16*	129.29
C_4	127.89		127.71	128.96
C_{11}	130.78	130.85	130.77	131.91
C_{12}	134.27	134.51	134.30	135.82
C_{13}	137.40*	137.42*	137.39*	138.61*
C_5	137.40*	137.52*	137.34*	138.42*
C_γ	128.07		128.06	
C_α	55.23	55.55	55.19	55.86
C_β	43.49	42.81	43.26	43.49
C_{Me}	26.67	26.51	26.61	28.02

[a] An asterisk indicates that these assignments could be reversed.

Structures A and D are invertomers in the T_{inv} process. Their separate existence has been demonstrated by the oxidation of cyproheptadine and the observation[25] of two isomeric *N*-oxides, both with NMe equatorial in the ratio of 3(D):1(A). The *N*-oxide D conformer is also a major metabolite detected in dogs fed cyproheptadine.[25]

From Figures 2 and 3 it is clear that the L and S conformers, now identified as D and A, respectively, are able to interconvert rapidly on the NMR time scale under certain conditions of temperature and solvent. Figure 1 illustrates that there are two possible mechanisms by which this interconversion may occur: combined P_{inv} and N_{inv} (which would ultimately depend on the rate of proton exchange) or T_{inv}. In many cases, NMR studies are not suited to distinguishing between mechanisms for dynamic processes, but in the current case the phase-sensitive NOESY spectrum offers a unique opportunity to define the process. In phase-sensitive mode, cross-peaks due to direct NOE effects appear as positive signals, whereas peaks arising from chemical exchange have a negative intensity. Consideration of the changes in environments for the various protons suggests that for the P_{inv}/N_{inv} process the chemical exchange peaks should appear as cross-peaks between the axial and equatorial hydrogens of the two conformers (e.g., H_{ae} in A exchanges with H_{aa} in D), whereas for T_{inv} the chemical-exchange cross-peaks should be found between axial hydrogens of the two conformers and between equatorial hydrogens of the two conformers. Figure 5a clearly shows that the former process leads to the observed interconversion between the L and S conformers. This conclusion is in agreement with the high experimental value (30.1 kcal/mol) for the interconversion barrier obtained from the racemization rates[26] for T_{inv} in 3-chlorocyproheptadine.

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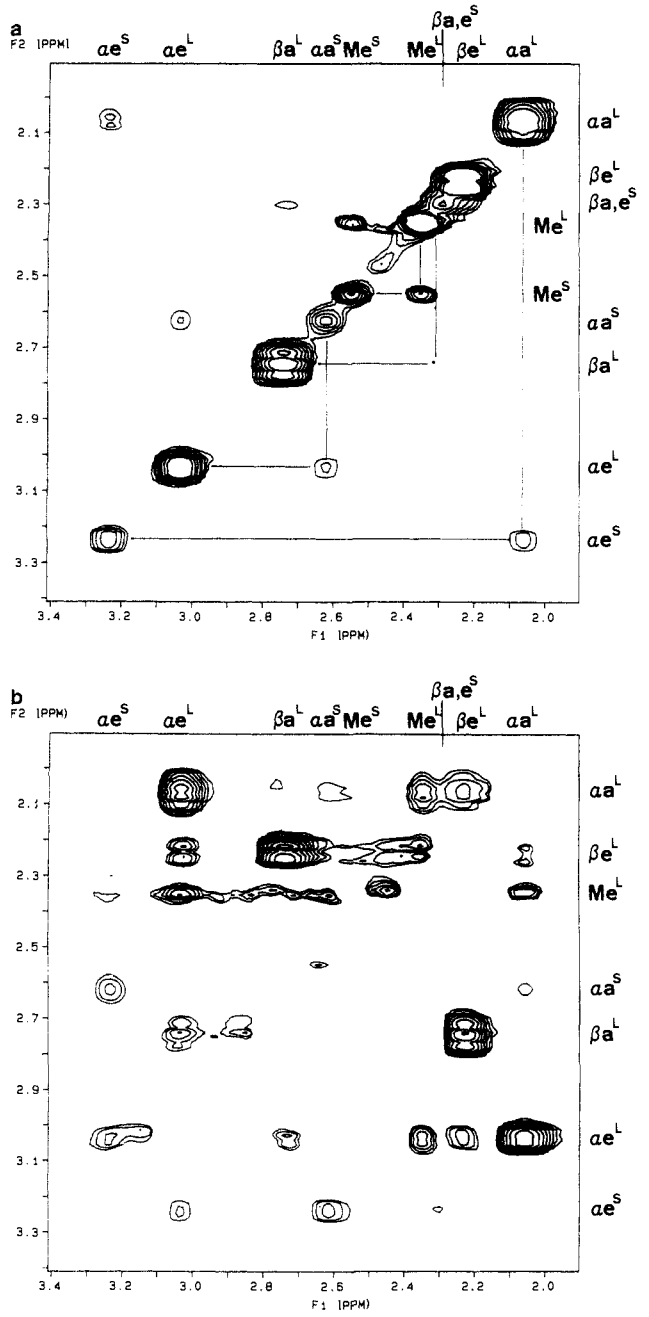


Figure 5. Aliphatic section of the same 2D NOESY spectrum as in Figure 4 showing (a) the negative diagonal and chemical-exchange peaks and (b) the positive NOE cross-peaks.

It is presumed that the same conformational processes for cyproheptadine hydrochloride are present in the ultrapure solvent. However, because of relatively fast proton dissociation/association rates, the lifetime of the N-protonated conformers is short on the NMR time scale, and only average shifts are recorded at room temperature. Chemical exchange peaks observed in 1D NOE difference experiments at low temperature confirm this presumption.

^{13}C resonances were found to behave in a similar fashion to the protons in the ^1H spectra discussed above. A single peak for each carbon is observed in ultrapure $CDCl_3$, and broadening of all resonances is observed on cooling of the solution. Insufficient resolution and freezing of the solution prevented an estimation of coalescence temperatures.

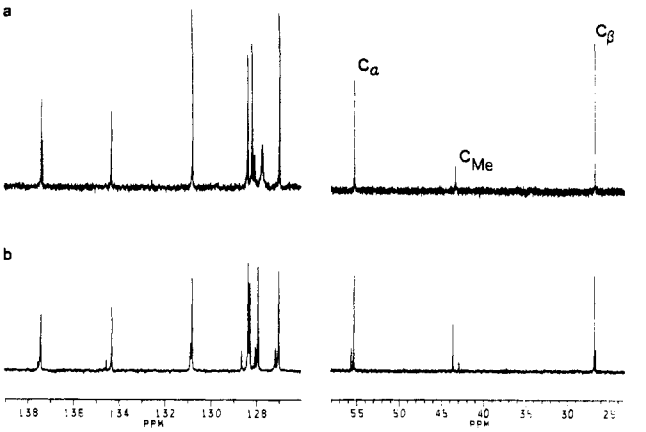


Figure 6. 300-MHz ^{13}C NMR spectrum of cyproheptadine hydrochloride in $CDCl_3$ at room temperature: (a) pure, (b) acidified.

Table III. Total Steric Energies (MM2) and Heats of Formation (AM1) (kcal/mol) Calculated for Four Conformations of Cyproheptadine

conformer	MM2 free base	AM1 free base	AM1 protonated
A	6.7	81.0	225.4
D	6.7	80.7	224.3
B	9.3	79.3	226.0
C	9.5	79.4	227.1

The assignments in Table II are based on additivity of substituent effects[27] and the 2D C–H correlation spectrum for the aromatic region recorded at room temperature. By contrast, the 1D ^{13}C spectrum of (1) in HCl-acidified $CDCl_3$ at room temperature clearly shows two shifts for each type of aliphatic carbon, with a peak height ratio of about 1:4 (Figure 6b). The aromatic region is more complex, but the resonances attributed to the more abundant conformer L are in agreement with the ^{13}C spectrum recorded in ultrapure $CDCl_3$ (Figure 6a).

The ^{13}C NMR spectrum of 1 in MeOD-d_3 at room temperature is similar to that discussed above. The suggested assignments are again presented in Table II. On lowering the temperature, splitting of all carbon resonances is observed, the ratio of conformers being close to 2:1 at 200 K. A barrier to the process of about 12.6 kcal/mol was estimated from the observed coalescence temperatures for aliphatic carbon resonances using the Eyring equation,[21] as discussed earlier.

^1H NMR spectra of 1 in MeOD-d_3 are more difficult to interpret than those in chloroform, although the aromatic region is very similar to that obtained in ultrapure chloroform at room temperature (Figure 2a). The aliphatic region at room temperature (Figure 7b) is complicated by the fact that all aliphatic peaks are close to coalescence. Lowering the temperature produced reversible spectral changes, but because the total aliphatic spin system was too large to allow total line-shape analysis using the DNMR-5 program[28] and available computing facilities were insufficient for expansion of the program, a quantitative explanation of the aliphatic region was not attempted.

III. Computational Study of Conformational Processes

An extensive theoretical study of conformational processes in cyproheptadine was carried out to provide con-

(26) Ebnother, A.; Jucker, E.; Stoll, A. *Helv. Chim. Acta* **1965**, *48*, 1287.
(27) Wehrli, F. N.; Wirthlin, T. *Interpretation of Carbon-13 NMR Spectra*; Heyden: London, 1978.
(28) QCPE Program 365; Quantum Chemistry Program Exchange, Bloomington, Indiana.

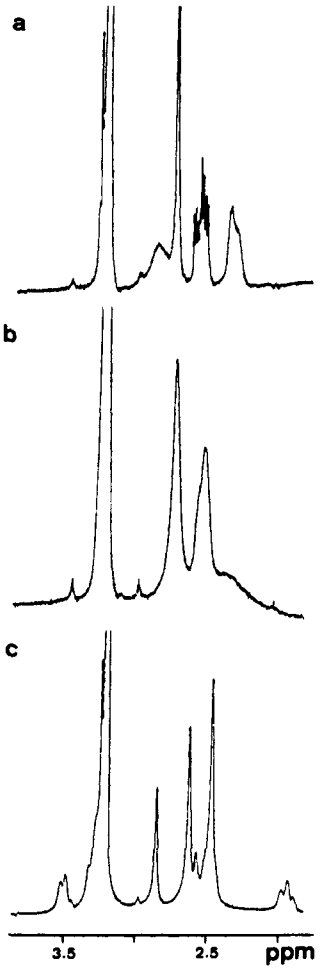


Figure 7. 300-MHz ^{1}H NMR spectrum of cyproheptadine hydrochloride in MeOD-d_3: aliphatic region at (a) 330 K, (b) 280 K, and (c) 200 K.

firmation for the above interpretation of the experimental NMR data. Of interest to us were the relative energies of the four possible conformers of cyproheptadine hydrochloride, and the energy barriers to the N_{inv}, P_{inv}, and T_{inv} processes.

Methods. The molecular mechanics program MM2[29] and the semiempirical molecular orbital (MO) package AMPAC,[30] with either AM1[31] or MNDO[32] parameterization, were used. For the MO calculations a standard Davidon–Fletcher–Powell geometry optimization procedure (supplied in the AMPAC package) was used except for the optimization of four protonated and unprotonated conformers of cyproheptadine (Table III). For these eight structures the AMPAC package was updated[33] to incorporate the Broyden–Fletcher–Goldfarb–Schanno geometry optimizer, which is more tolerant of rounding-off errors and converges faster than the standard procedures. Structures A and D, as free bases, were calculated by both methods starting from the same input files. However, the results did not produce clear evidence for the preference of one method over the other.[34]

A simple methodology involving systematic stepwise changes in a single torsion angle and optimization of the remaining structural parameters at each step is usually applied for the estimation of rotational barriers. This method is, however, unreliable for the determination of conformational barriers in ring structures.[35] Because of the interdependence of torsion angles in the ring and the existence of multiple optimum values for the full set of ring torsion angles following variation of one of them, this method may not reveal the true inversion pathway.

One solution to this problem is to compute multidimensional energy surfaces and inspect them for available minima and saddle points that correspond to transition states. However, the enormous increase in computational time necessary to calculate surfaces renders this solution impracticable for most compounds. Thus, the conformational barrier is usually estimated as an energy difference between the *postulated* transition state and the optimized ground states.[36-38] Extreme care should then be taken in interpreting any agreement between the experimental and calculated values.

A more exact calculation of the energy barrier would require the calculation of energy changes associated with continuous deformation of the geometry across a pathway from one minimum (ground state) to another, passing through the lowest possible saddle points. The probability of correctly guessing such minimum-energy pathways decreases significantly with the increasing complexity of the structure (and hence the complexity of the conformational energy surface); thus only estimates of barriers may be obtained by investigating one or a few nominated pathways and by searching and examining transition states in their vicinity.

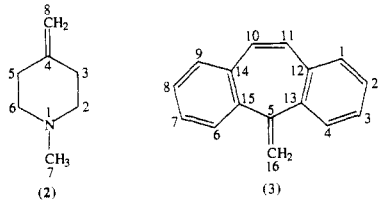
To estimate the energy barrier to ring and nitrogen inversion in cyproheptadine, the reaction-coordinate ap-

(29) Program MM2(85); Molecular Design Ltd., 2132 Farallon Drive, San Leandro, CA 94577.
(30) Dewar, M. J. S.; Stewart, J. J. P. *QCPE Bull.* **1986**, *6*, 24 (QCPE Program 506).
(31) Dewar, M. J. S.; Zoebisch, E. V.; Healy, E. F.; Stewart, J. J. P. *J. Am. Chem. Soc.* **1985**, *107*, 3902.
(32) Dewar, M. J. S.; Thiel, W. *J. Am. Chem. Soc.* **1977**, *99*, 4899, 4907. Dewar, M. J. S. *J. Phys. Chem.* **1975**, *89*, 2145.
(33) Cioslowki, J.; Kertesz, M. *QCPE Bull.* **1987**, *7*, 159.
(34) The results from these four calculations indicate that any CPU time saving is obtained only in the case of faster convergence, i.e., exiting after a lower number of cycles. However, because the AMPAC package has several routes (criteria) for the termination of optimizations (as described in the AMPAC manual[30]) and there is no provision for forcing exit via a selected criterion, the use of the new optimization method may actually lead to an increase in CPU time, as happens for conformation D. Thus in a comparison of energies for different conformations optimized under the same conditions but happening to exit the calculation via different routes, an additional factor may bias the results. Generally, the more cycles the optimization goes through, the lower the heats of formation ΔH and the lower the gradient norm ΔG, but the longer the CPU time. We did not reoptimize these four structures using PRECISE (AMPAC manual[30]) because, in our experience, this does not guarantee solution of the problem, and sometimes leads to a considerable increase in CPU time, especially for the optimization of large molecules. The final two A conformations and the two D conformations have only a small difference in energies and small differences in geometries (less than 0.01 Å for bonds, less than 0.1 Å for nonbonded distances, and approximately 2° for bond angles). These minor differences are considered insignificant for modeling purposes.

	A-DFP	A-BFGS	D-DFP	D-BFGS
ΔH, kcal/mol	80.90	81.04	80.75	80.68
ΔG, kcal/mol per Å	4.80	7.13	6.12	2.62
cycles	64	32	32	40
SCF	281	138	138	178
exit[30]	ΔH	Herbert	Herbert	ΔH
CPU time, min	737	383	378	480

(35) Burkert, U.; Allinger, N. L. *J. Comput. Chem.* **1982**, *3*, 40.
(36) Downing, A. P.; Ollis, W. D.; Nogradi, M.; Sutherland, I. O. *Jerusalem Symp. Quant. Chem. Biochem.* **1971**, *3*, 296.
(37) Nogradi, M.; Ollis, W. D.; Sutherland, I. O. *Chem. Commun.* **1970**, 158.
(38) Lambert, J. B.; Featherman, S. I. *Chem. Rev.* **1975**, *75*, 611.

proach was applied with the structures of 1-methyl-4-methylenepiperidine (2) and 5*H*-dibenzo[*a*,*d*]cyclohepten-5-ylidene (3). As a starting point, N-Me_{ax} and



(2) (3)

N-Me_{eq} chair conformations of 2 and the boat conformation of 3 were built from fragments within the CRYSX[39] graphics program. The energies along pathways were obtained with semiempirical calculations, as they allowed comparison between inversions of the free-base piperidine ring and the protonated piperidine ring. Molecular mechanics calculations, which are less demanding on computational time but also less suited to studying charged or highly conjugated compounds,[40] were also used in some instances. The most recent AM1 parameterization[31] available in the AMPAC package was chosen. Some of the transition states along the pathways were located with the saddle-point calculations of Dewar[41] and were confirmed as true transition states by the presence of only one negative eigenvalue in their force constant (Hessian) matrices. Plots of conformational energy surfaces were produced by the program ENMAP.[42]

Results and Discussion. (i) Relative Energies of Stable Conformers of Cyproheptadine. The energies of the four cyproheptadine conformations A–D, either as free bases (MM2, AM1) or as protonated cations (AM1), are summarized in Table III. Conformation A was indicated by MM2 to be less stable than conformation D; however, the difference is not considered significant. The N-Me_{ax} conformations, B and C, were about 2.6–2.8 kcal/mol less stable than N-Me_{eq} conformations A and D. These results support N-Me_{ax} conformers being nonobservable by NMR, although the conformational pathway suggests their, at least transient, existence. The energy difference between the axial and equatorial conformers is similar to that found in 1-methylpiperidine.[43]

The semiempirical calculations do not reflect the trend shown by MM2. They indicate the axial conformers B and C to be about 1.5 kcal/mol more stable than the equatorial conformers A and D. The calculated energies of the N-protonated conformers appear to be in the relative order expected from experimental data on cyproheptadine and similar compounds already discussed. Thus it seems that the AM1 parameterization cannot handle the repulsive interactions of the nitrogen lone pairs and hydrogens in the axial positions correctly.

No matter what calculations were used, the geometries of A,D and B,C were very similar except for the position of the NMe group. The X-ray structure of cyproheptadine[14] is far less symmetrical than the optimized structures, probably the result of packing. Protonation has no significant effect on the overall geometry of the optimized

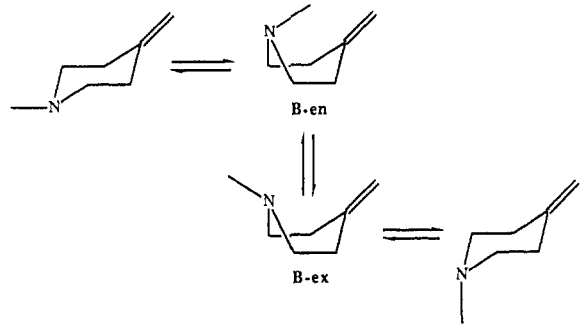


Figure 8. Inversion of the piperidine ring of 2 showing the two possible boat transition states, B-en and B-ex.

structures, characterized by the distance of the nitrogen from the planes of benzene rings, the angle between these two planes, the distance of the nitrogen from the ethylene bridge, and the boat shape of the central heptatriene ring. According to the same criteria, there is no significant difference between the AM1 and MM2 geometries except for the angle between the two benzene planes, which is 5° more open in the latter structures. From a comparison of the AM1-optimized protonated conformations with the X-ray structure,[14] it is obvious that the latter agrees with conformation D. However, the X-ray structure has a flatter boat conformation for the central ring and therefore a 0.4 Å shorter distance between the nitrogen and the benzene planes and a 0.4 Å longer distance between the nitrogen and the ethylene bridge. The angle between the benzene planes is about 2° smaller than in the AM1-optimized protonated conformation D.

(ii) Piperidine Ring Inversion. The inversion pathways of 1-methyl-4-methylenepiperidine (2, chair conformation) are asymmetrical, with two possible boat transition states (possibly intermediates), B-en and B-ex (Figure 8), related either by pseudorotation with numerous transition states/intermediates or by nitrogen inversion (discussed later), all of which indicate a multitude of possible transition states and local minima. The prediction of a minimum-energy pathway and estimation of the barrier to inversion by reaction coordinate calculations without studying all pathways are therefore a matter of guesswork.

Figure 9 illustrates some of the possible pathways and transition states studied by driving the N_1–C_2–C_3–C_4 torsion angle and optimizing (using AMPAC) all other remaining geometrical parameters at each step, with or without enforcement of symmetry along the N_1–C_4 axis. The AM1 parameterization indicates that the N-Me_{eq} conformer of unprotonated compound 2 is 1.5 kcal/mol higher in energy compared with the N-Me_{ax} conformer, contrary to expectation. In protonated compound 2 the energy for the N-Me_{eq} conformer was initially found to be 0.2 kcal/mol below that of the axial conformer. Using either a different starting structure or the same structure with a different geometry definition in terms of the internal coordinates (an infinite number of possibilities is available), another N-Me_{eq} conformer was found 1.5 kcal/mol below the N-Me_{ax} conformer. There are two points worthy of emphasis: (a) the only significant difference between these two equatorial conformers is that the lower energy one is one degree closer to the true sp^3 configuration around the nitrogen, and (b) we did not succeed in obtaining this lower energy structure by continuing to optimize the higher energy conformer with stricter convergence limits (AMPAC manual[30]). This strategy, successful in locating the low-energy protonated N-Me_{eq} conformer of 2, failed to find the lower energy structure for the unprotonated N-Me_{eq} conformer of 2. Thus it seems that the energy difference

(39) Sadek, M.; Munro, S. L. A. *J. Comput.-Aided Mol. Des.* **1988**, *2*, 81.
(40) Clark, T. *A Handbook of Computational Chemistry*; Wiley Interscience: New York, 1985.
(41) Dewar, M. J. S.; Healy, E. F.; Stewart, J. J. P. *J. Chem. Soc., Faraday Trans. 2* **1984**, *80*, 227.
(42) Program ENMAP based on subroutine KONTOR. Palmer, J. A. B. *Aust. Comput. J.* **1970**, *2*, 27.
(43) Katritzky, A. R.; Patel, R. C.; Riddell, F. G. *Angew. Chem., Int. Ed. Engl.* **1981**, *20*, 521.

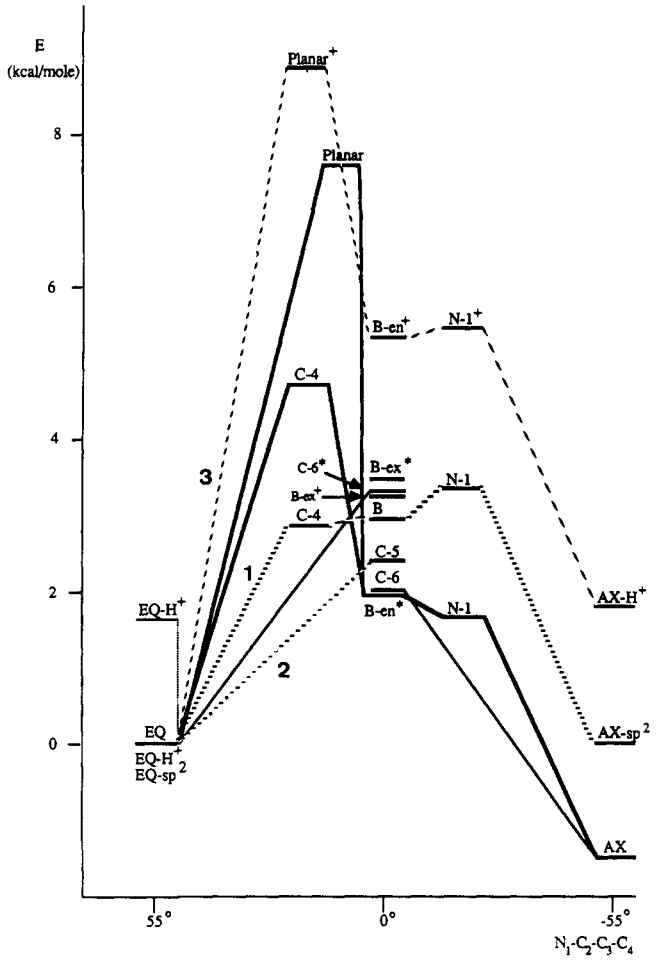


Figure 9. Relative energies (all N-Me_{eq} set to zero) of located minimum conformations and some characterized transition states (*) on some of the interconversion pathways investigated for compound **2**: thick line represents unprotonated **2** calculated with symmetry; thin line represents unprotonated **2** calculated with no symmetry, both with nitrogens in sp^3 configuration; broken line 1 represents unprotonated **2** calculated with symmetry; broken line 2 represents unprotonated **2** calculated with no symmetry, both with nitrogens in sp^2 configuration; and broken line 3 represents protonated **2** calculated with symmetry. On the left and right are minima corresponding to the chair conformations, with N-Me_{eq} and N-Me_{ax}, respectively; in the middle are mainly boat conformations. The remaining conformations have five ring atoms coplanar and are designated by the atom that is out of this plane.

between the equatorial and axial conformers of **2**, calculated by AM1 parameterization, is genuine and can be ascribed to the overestimated repulsion of the N-axial lone pairs and axial ring hydrogens discussed above.

Optimization of these four structures was repeated with MNDO parameterization[32] and the relative energies were, qualitatively, as expected; i.e., N-Me_{eq} conformer (free base) was 1.3 kcal/mol below the axial conformer and the N-Me_{eq} conformer (protonated) was 1.4 kcal/mol below the protonated axial conformer. However, we did not think this warranted recalculation of all the data with MNDO parameterization.

Further inspection of Figure 9 indicates that the inversion paths through distortion of the configuration around the nitrogen (flipping the N_1 vertex of the ring) are energetically more demanding and that the actual path is sensitive to the starting point and direction taken along the coordinate (i.e., the path is not retraceable by changing the direction). The relative rigidity of the N_1 side of the ring is in agreement with the 2 kcal/mol higher inversion barrier in *N*-methylpiperidine[23b,c] compared with that in cyclohexane[44,45] and a similar decrease in the inversion barrier for 1-methylenecyclohexane.[46-48] With a single reaction coordinate it is possible to pass from the equatorial to the axial conformation only by enforcing the symmetry of the molecule at each step. Without symmetry enforcement, the closest transition state on the pseudorotational surface is reached, and continuation along the same coordinate finishes at another transition state and discontinuity. By forcing the nitrogen to remain in an sp^2 (planar) geometry (the transition state for the N_{inv} process is discussed below) the inversional barriers are modified, confirming the interdependence of the P_{inv} and N_{inv} processes. This reaction pathway, contrary to normal expectation, is still asymmetrical, illustrating perfectly the lagging effect and entrapment in side valleys of an unknown energy surface, which has been discussed in detail by Burkett and Allinger.[35] The same effect is also illustrated by the differences in pathways between the equatorial conformer and the boat transition state, and in pathways with and without enforcement of molecular symmetry.

A number of boat B-en transition states were located with symmetrical or skewed boat geometries (up to 19° for the N_1–C_5–C_6–C_4 torsion angle); all were within 0.5 kcal/mol (only one is shown in Figure 9). They were characterized as true transition states by a single negative eigenvalue in their Hessian matrices.[41] The B-ex transition state was found to be 1.5–2.0 kcal/mol higher in energy, presumably because of repulsion between the lone pair of the nitrogen atom and the π-orbitals of the exocyclic double bond. The same magnitude of difference is indicated between the boat transition states of the protonated conformers, but here it is the B-ex that is lower in energy, presumably due to stabilization by favorable proton and π-orbital interactions. Apart from that, the interconversion pathways for the protonated conformers of **2** seem analogous with those for the unprotonated conformers, with about 50% higher interconversion barriers.

The estimated barriers (5.0–3.5 kcal/mol for the axial to B-en and 3.5–2.0 kcal/mol for the equatorial to B-en inversions, Figure 9) are rather low compared with the experimentally derived inversion barriers in simple *N*-methylpiperidines (10–12 kcal/mol[23b,44]) and 1-methylenecyclohexane (7.7 kcal/mol[46-48]), suggesting that AM1 parameterization severely underestimates the inversion barriers of six-membered rings. Hence inversion of the piperidine ring of **2** was also investigated with the MM2 program[31] with no alteration to the force field.[49] The nitrogen atom was characterized as type 8 (according to the nomenclature used within the MM2 program), with the specific inclusion of a lone pair. The inversion surface in Figure 10 was obtained by simultaneous driving of two torsion angles, N_1–C_2–C_3–C_4 and N_1–C_6–C_5–C_4, in 10° steps starting from the boat structure B-en. It is satisfying to note that the energy profile along the diagonal connecting the N-Me_{ax} and N-Me_{eq} conformer minima is qualitatively the same and passes through the same geometries, as the reaction coordinate along the N_1–C_2–C_3–C_4 torsion angle, calculated with symmetry enforcement by AM1 paramet-

(44) Harris, R. K.; Spragg, R. A. *Chem. Commun.* **1966**, 314.
(45) Anet, F. A. L.; Bourn, A. J. R. *J. Am. Chem. Soc.* **1967**, *89*, 760.
(46) Gerig, J. T.; Ortiz, C. E. *J. Am. Chem. Soc.* **1970**, *92*, 7121.
(47) Gerig, J. T. *J. Am. Chem. Soc.* **1968**, *90*, 1065.
(48) Jensen, F. R.; Beck, B. H. *J. Am. Chem. Soc.* **1968**, *90*, 1066.
(49) Allinger, N. L. *J. Am. Chem. Soc.* **1977**, *99*, 8127. Allinger, N. L.; Flanagan, H. L. *J. Comput. Chem.* **1983**, *4*, 399. Current parameter set obtained through private communication from N. L. Allinger.

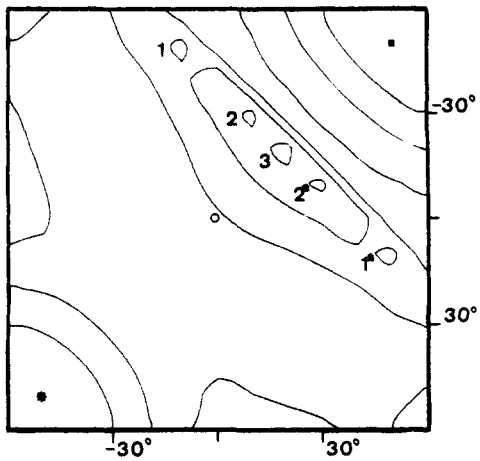


Figure 10. Conformational energy surface for **2** obtained by systematic stepping of torsion angles C_4–C_3–C_2–N_1 (horizontal axis) and C_4–C_5–C_5–N_1 (vertical axis) starting from the middle of the map. The contours are at 2.5 kcal/mol intervals: * AX (11.4 kcal/mol), ■ EQ (9.0 kcal/mol) ○ B-en (18.9 kcal/mol). The maxima 1 and 1* are 22.1 kcal/mol (C_6–C_3 and C_5–C_2 boats), 2 and 2* are 24.5 kcal/mol (flat B-en), and 3 is 25.0 kcal/mol (only C_4 out of plane of other ring atoms).

erization (Figure 9). In better agreement with the experimental data mentioned above, the N-Me_{ax} conformer is 2.4 kcal/mol less stable than the N-Me_{eq} conformer, and the inversion barrier values of 7.5 and 9.9 kcal/mol are more realistic than the AM1 results. However, the B-ex boat structure is now 3.1 kcal/mol more stable than the B-en boat structure. Thus, although the barriers do not seem to be as underestimated as in the case of the semiempirical MO calculations, their ability to account for the electronic interactions is limited.

Neither molecular mechanics nor the semiempirical MO method give entirely satisfactory results, nor is the reaction coordinate approach very efficient, often allowing only a qualitative assessment of the situation. In more complex situations, for example, in cyproheptadine itself, the number of possible boat transition states and structures on the pseudorotational surface would be doubled because of the two possible orientations of the tricyclic ring, which replaces the symmetrical 4-methylene substituent.

(iii) Nitrogen Inversion. Compared with piperidine ring inversion, the reaction coordinate (the geometry changes along the inversion pathway) for N_{inv} is unambiguous. The process was again modeled on 1-methylene-4-piperidine (**2**) and it was thus necessary to consider only one transition state for the chair conformer of **2**, that with sp^2 geometry around the nitrogen. The barrier of 6.0 and 4.5 kcal/mol (AMPAC, AM1 parameterization) was obtained by driving the torsion angle C_6–C_2–N_1–C_7 from 120° (the N-Me_{ax} conformer) to 240° (the N-Me_{eq} conformer). The pathway is retraceable, going from the equatorial back to the axial conformer. The barrier is comparable with that of 6.0 kcal/mol, derived from an ultrasonic relaxation study,[50] for the N-Me_{ax} to transition-state inversion of 1-methylpiperidine. The angle C_6–N_1–C_5 opens by 8° on passing through the transition state, and the dihedral angles within the ring undergo up to 11° of change along the pathway. The structures along the pathway are symmetrical to within 1°, and several transition states with very similar geometry to sp^2 geometry around the nitrogen (the C_6–C_5–N_1–C_7 torsion angle = 180° ± 1°) were confirmed by the presence of single negative

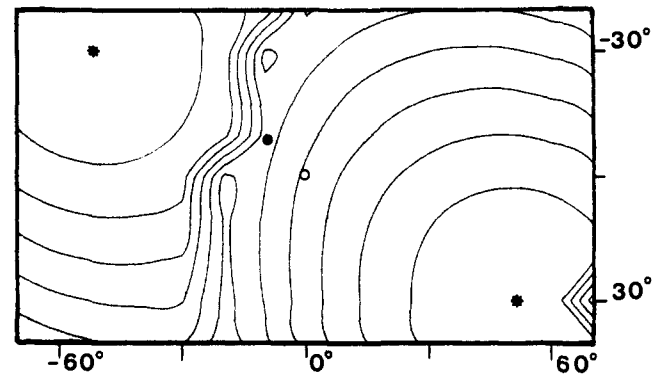


Figure 11. Conformational energy surface for **3** obtained by systematic stepping of torsion angles C_{14}–C_{15}–C_5–C_{13} (horizontal axis) and C_{13}–C_{12}–C_{11}–C_{10} (vertical axis) starting from * boat conformation B-1 (-4.5 kcal/mol). In compound 3 conformations B-1 and B-2 are identical; ● is conformation T-1 (9.0 kcal/mol) and ○ is conformation T-2 (4.9 kcal/mol) described in the text. Contours are at 2.5 kcal/mol intervals.

eigenvalues in their Hessian matrices. Following the segments of the N_{inv} pathway between 120° and 170° and 190° and 240° with the protonated chair conformer of **2** indicates that resistance to geometry flattening around the nitrogen increases 5–6 times. The structures along the pathway deviate from symmetry by up to 5°, the largest change observed in dihedrals being 17°.

(iv) Inversion of the Tricyclic Ring. Inversion of the tricyclic ring of cyproheptadine was modeled on the structure of 5*H*-dibenzo[*a*,*d*]cyclohepten-5-ylidene (**3**). The energy surface (Figure 11) representing inversion of the central seven-membered ring in **3** was obtained with the MM2 program with special parameters for benzene rings.[51] Although it was not expected that MM2 would give an accurate account of the electronic interactions in this highly unsaturated molecule, the steric interactions are quite well illustrated by this map. The two B-1 and B-2 boat invertomers are identical and fully superimposable. Note that only the fully planar transition state is achiral, even in the case of no substitution at the tricyclic ring. The surface in Figure 11 was obtained by stepwise driving of two chosen angles, C_{14}–C_{15}–C_5–C_{13} and C_{13}–C_{12}–C_{11}–C_{10}. Lack of symmetry in the surface may indicate asymmetrical distribution of strain as the molecule passes through the transition state, which need not be symmetrical either. Neither the highest saddle point, T-1, nor the center of the map, T-2, has planar geometry. According to this map the barrier to inversion is 13.5 kcal/mol. Force calculations identified both T-1 and T-2 as local minima only. In an attempt to optimize the structure into a fully planar one by applying strict symmetry along the mirror plane passing through the exocyclic double bond and the middle of the ethylene bridge and by fixing the torsion angles C_{14}–C_{15}–C_5–C_{13} and C_{15}–C_{14}–C_{10}–C_{11} to zero, the structure T-3 was obtained. This has a planar seven-membered ring. However, the benzene rings are still in a butterfly conformation and the exocyclic double bond is also out of plane but trans to the benzene rings. This conformation was also identified as a local minimum only 11.6 kcal/mol above the boat conformation.

Although the transition state was not found, the calculations at least indicate that the fully planar structure is excessively strained and unoptimizable without severe geometry restrictions. A reasonable strategy for location of the transition state may be a fine grid search (1° steps) in the vicinity of the T-1 structure. However, we were

(50) Gittins, V. M.; Heywood, P. J.; Wyn-Jones, E. *J. Chem. Soc., Perkin Trans. 2* **1975**, 1642.

(51) Allinger, N. L. *QCPE Bull.* **1983**, *3*, 32.

more interested to see the effect of the extension of 3 into a full cyproheptadine on the T inversion barrier. The A and D cyproheptadine conformations were built with T-3 for the tricyclic nucleus and reoptimized by using the same geometry restrictions as for T-3. Considering these new optimized structures as approximations to the transition state, barriers of 28 kcal/mol with MM2 and 17 kcal/mol with AM1 were obtained. Thus, with regard to the available experimental data (this paper and ref 26), it can be concluded that the energy values calculated by AM1 are misleading.

IV. Conclusions

It has been shown that cyproheptadine hydrochloride is relatively flexible in solution, with four accessible stable conformers (A, B, C, and D). Of these, A and D were observed experimentally by NMR. Evidence for the existence of B and C was inferred from the suggested conversional mechanism, for which there is support from the 2D NOESY spectrum. Two of the conformational processes discussed P_{inv} and N_{inv}, have been shown to be sensitive to the acidity of the solution.

The computational methods MM2 and AM1 have been found to be deficient in estimating the energy barriers to the conformational processes studied. The deficiencies of the AM1 method observed are in agreement with those recently reported[52,53] for other structures. It is noted that irrespective of the computational method used, the skeleton geometry of the molecules in related conformations remains, within small tolerances, the same. For cyproheptadine this means that the geometries (apart from the orientation of the NMe moiety) of conformations A and B are the same as those of D and C, respectively. This is probably the result of parameterization at both levels, MM and MO, based on the geometry of the X-ray-analyzed structures. Thus the choice of method becomes critical only when information on relative energies is required together with the plausible geometry of a compound.

It has been established that the nitrogen and aromatic ring is a core pharmacophore for most CNS-active compounds.[54] The selectivity of these is then presumably mediated through the secondary binding groups and by the fine differences between the relative positioning of the relevant nitrogen and aromatic ring. It has been further suggested that the optimal distance for the protonated nitrogen from the plane (or centroid) of the relevant aromatic ring is about 4 Å (5–5.5 Å) for tricyclic antidepressants, around 5 Å (around 6 Å) for antimuscarinics, and around 1.5 Å (around 6 Å) for the neuroleptics.[5] Note that there is no direct relationship between the distances of the active nitrogen to the plane of the aromatic ring and the nitrogen to the centroid of that ring. Understandably there would also be other parameters determining the selectivity apart from the primary requirements discussed above.

Borea et al.[15] derived an H-1 antihistaminergic model, based on 14 H-1 antagonists, where the crucial parameters seem to be the distance between the nitrogen and the centroid of the aromatic plane (6.2 ± 0.15 Å) and the distance between the centroids of the two aromatic rings (4.9 ± 0.24 Å) found in these antihistaminergics. The X-ray conformation of cyproheptadine hydrochloride[14] fits this model as such. Essentially the same model was described by Naruto et al.,[16] with a nitrogen to centroid distance of 6.5 and 5.8 Å and the distance between the centroids of 4.9 Å. The conformation of cyproheptadine when fitted to this model is slightly asymmetrical and about 1.7 kcal/mol above the minimum conformation (presumably D). It is notable that the same distance between the nitrogen and the centroid of the aromatic ring is quoted[5,15] for all antimuscarinics, neuroleptics, and antihistaminergics.

In the AM1-calculated D,B conformations (for both protonated and unprotonated cyproheptadine), the distance from the nitrogen to the plane of the aromatic ring is 3.8 ± 0.2 Å (3.4 ± 0.1 Å) in the X-ray structure[14]), and the distance between the nitrogen and the centroid of the ring is 6.2 ± 0.1 Å (6.28 ± 0.03 Å in the X-ray structure). In the A,C conformations the values are 2.7 ± 0.2 and 6.2 ± 0.1 Å, respectively. These parameters seem to be (except for the antihistaminergics) outside the ranges quoted above for drug classes containing the tricyclic nucleus. Accepting the D conformer as that responsible for H-1 activity would nominate the A conformer as the 5HT-2 agent. In terms of the known 5HT-2 activity of some tricyclic antidepressants (e.g., mianserin[55]), this may be controversial. Considering the almost equipotent activity of cyproheptadine at the H-1, M-1, and 5HT-2 receptors, it would seem that either there is a greater overlap of the two major geometrical parameters discussed or the active conformations are other than the stable conformations A and D.

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Registry No. 1, 129-03-3; 1·HCl, 969-33-5; 1·H$^+$, 124993-56-2; 2, 13669-28-8; 2·H$^+$, 124993-57-3; 3, 2975-79-3.

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