Conformational Analysis of Cyclic Amines using Carbon-13 Chemical Shift Measurements: Dependence of Conformation upon Ionisation State and Solvent.

By John G. Batchelor, National Institute for Medical Research, Mill Hill, London NW7 1AA

Using observations on model compounds a set of substituent shift parameters has been derived which can be used to predict the ^{13}C chemical shifts and protonation shifts of the conformers of cyclohexylamine derivatives. Large differences are predicted between the spectra of the different conformers and, therefore, the conformational equilibria of these compounds can be determined from the observed chemical shifts of their ^{13}C resonances. The limit on the accuracy of the method is at present ±2% for conformers with small conformational free-energy differences but may be further improved by increased understanding of ^{13}C chemical shifts. The present accuracy is sufficient to enable the effects of ionisation state and solvent on the conformation to be investigated. An increase in ΔG of 1.6 kJ mol^{-1} is observed upon changing from cyclohexane to D_2O as solvent and a further increase of 1.2 kJ mol^{-1} is measured upon protonation of the NH_2 group.

METHODS employing n.m.r. have played an important part in attempts to determine the conformational equilibria of cyclohexane derivatives.[1-6] The most accurate method for measuring the free-energy differences between conformers, ΔG, is the direct measurement at low temperature of the areas of methine ^1H resonances corresponding to the separate conformers.[3] However, this method has limited usefulness for estimating conformational equilibria at ambient temperature because ΔG is dependent on temperature, through an entropy contribution, and ΔG values can only be determined in solvents with freezing points lower than *ca.* −100 °C. Thus it is not possible to determine conformations in aqueous solutions nor to investigate the solvent dependence of conformational free energies, a very significant dependence for compounds such as amines.[4] For such purposes it is necessary to investigate the spectra under the solvent and temperature conditions of interest.

At ambient temperatures the different conformers of cyclohexane derivatives are in fast exchange, so that for each nucleus only a single resonance is observed with average shielding and couplings dependent on the time spent in each conformer. Since it is not possible to measure directly the spectral characteristics of each conformer a major problem in conformational analysis using spectra recorded in the fast-exchange limit is the estimation of these characteristics. For example, chemical shifts have been estimated either by extrapolation from shifts of the individual conformers observed at low temperature [1] or by comparison with other molecules with well defined conformation.[2] The first method suffers from the problem of estimating the temperature dependence of the shifts and the restricted range of solvents which are liquid at low temperature. In the second method it has been necessary to assume that 4-t-butyl groups, while locking a molecule into a single conformation, have no effect on the methine ^1H chemical shifts. However, this assumption has proved inadequate.[7]

The other major problem encountered in such studies at ambient temperature using ^1H n.m.r. is that of spectral resolution. The only chemical shift which can easily be measured is that of the methine ^1H and, to measure ^1H—^1H couplings, it is often necessary to use selective decoupling techniques and isotopic enrichment with deuterium.

Carbon-13 chemical shifts possess a number of advantages which may make them a useful tool for investigating conformations at ambient temperatures.[8] Proton noise-decoupled ^{13}C spectra contain sharp, well resolved resonances so it is generally possible to measure accurate chemical shifts for all the carbons in small compounds. Thus several independent determinations of the conformation may be made, thereby increasing the accuracy of the final result and also providing an estimate of its reliability. The changes in chemical shift of ^{13}C resonances in cyclic systems occurring upon ring inversion, the 'conformational shifts', are large, generally in the range 3—7 p.p.m., and thus can be measured very accurately (± 0.02 p.p.m. in this study) so that experimental contributions to errors in measuring conformation will be $< 1\%$. Because of the high resolution of ^{13}C chemical shifts it is also possible to assign the resonances and determine the conformations of molecules in mixtures of geometric isomers without separating them, as demonstrated here.

In ^{13}C n.m.r. it is impossible to ignore the effects of a substituent on the resonance of any carbon within the ring. However, it has been shown for methyl substituents that, using the ^{13}C shifts of compounds with well defined conformations, a set of additive substituent parameters can be developed to predict accurately the chemical shifts of all ring resonances in conformationally homogeneous compounds with a standard error of < 0.3 p.p.m.[9,10] Thus from the chemical shifts of each

[1] H. Booth, *Tetrahedron*, 1964, **20**, 2211.
[2] E. L. Eliel and R. J. L. Martin, *J. Amer. Chem. Soc.*, 1968, **90**, 682.
[3] F. R. Jensen, C. H. Bushweller, and B. H. Beck, *J. Amer. Chem. Soc.*, 1969, **91**, 344.
[4] H. Booth in *Progr. N.M.R. Spectroscopy*, 1969, **5**, 149.
[5] O. A. Subbotin and N. M. Sergeyev, *J. Amer. Chem. Soc.*, 1975, **97**, 1080.
[6] H. Booth, *J. Chem. Soc. Chem. Comm.*, 1973, 945.
[7] F. R. Jensen and B. H. Beck, *J. Amer. Chem. Soc.*, 1968, **90**, 325.
[8] J. B. Stothers, 'Carbon-13 NMR Spectroscopy,' Academic Press, New York, 1972.
[9] D. K. Dalling and D. M. Grant, *J. Amer. Chem. Soc.*, 1967, **89**, 6612.
[10] D. K. Dalling and D. M. Grant, *J. Amer. Chem. Soc.*, 1972, **94**, 5318.

individual ^{13}C resonance it should be possible to estimate molecular conformation to better than ± 10%.

The purpose of this article is, first, to derive a set of parameters for predicting the shifts of cyclic amines and then to show that it is possible to estimate the conformations of cyclic amines without purification of isomers, and to show how these conformations change upon protonation and upon changing from protic to aprotic solvent systems.

EXPERIMENTAL

Cyclohexylamine and the methylcyclohexylamines used were all obtained commercially and used without further purification. The dimethylcyclohexylamine sample was kindly donated by Dr. H. Booth. Aqueous solutions were prepared containing 1 : 2 : 6 parts by volume of sample, dioxan, and D_2O. ^{13}C Spectra were recorded for solutions adjusted to two or more pH units above and below each pK value using concentrated DCl or KOD. In general it was also necessary to record spectra at intermediate pH values to determine the correspondence of the resonances in protonated and unprotonated molecules. Nonpolar samples were prepared 25% by volume in cyclohexane.

Carbon-13 proton noise-decoupled spectra were obtained on a Varian XL-100-15 spectrometer operating at 25.2 MHz in the Fourier transform mode using 8K data sets. A 2 000 Hz spectral width was used providing chemical-shift measurements correct to ± 0.02 p.p.m. Dioxan and cyclohexane were used as internal references and chemical shifts were converted to the $SiMe_4$ (TMS) scale using the relationships

$$\delta^C_{TMS} = \delta^C_{dioxan} + 67.3 = \delta^C_{C_6H_{12}} + 27.4 = \delta^C_{C_6H_6} + 128.8$$

RESULTS

Assignments.—Observations were made on five samples; cyclohexylamine, and mixtures of *cis*- and *trans*-2-methyl-, 3-methyl-, 4-methyl- and 3,5-dimethyl-cyclohexylamines (Table 1). The assignments of cyclohexylamine have been made previously.[11] For the remaining compounds the assignments of the conformationally homogeneous isomers were made first. Then accurate substituent shift parameters derived from a comparison of the chemical shifts of all the unequivocally assigned resonances with the chemical shifts of their parent alkane resonances [10] were used to assign the conformationally heterogeneous isomers. For 4-methyl- and 3,5-dimethyl-cyclohexylamines the resonances of the isomers could be differentiated by their large differences in intensity. The assignments of the conformationally homogeneous isomers (*trans*-, and *cis*-, and *trans*-, respectively) were then made by analogy with the ^{13}C spectra of cyclohexylamine and the parent alkanes.[10] A preliminary set of amino-substituent shift parameters was then derived. These parameters were used to assign, in the spectra of 3-methyl- and 2-methyl-cyclohexylamine, the resonances of the isomers in which the diequatorial conformer dominates (*cis*- and *trans*-isomers, respectively). All definite assignments made on the basis of such chemical-shift comparisons were supported by the shifts of the resonances observed upon protonation of the amino-group in D_2O.[12]

The chemical shifts of the chair conformers with axial and equatorial amino-groups, of *cis*-4-, *trans*-3-, and *cis*-2-methyl cyclohexylamines were then calculated and plotted as illustrated in the Figure. The vertical lines represent the shifts of the observed resonances. Only for one small range of conformations is there close agreement between the observed and predicted shieldings of the ring carbons, this range being consistent with the conformations expected on the basis of published free-energy values for amino- and methyl groups.[4] Thus the resonances could be assigned and their chemical shifts used in the calculation of the molecular conformation.

TABLE 1

Carbon-13 chemical shifts [a] of cyclohexylamine derivatives

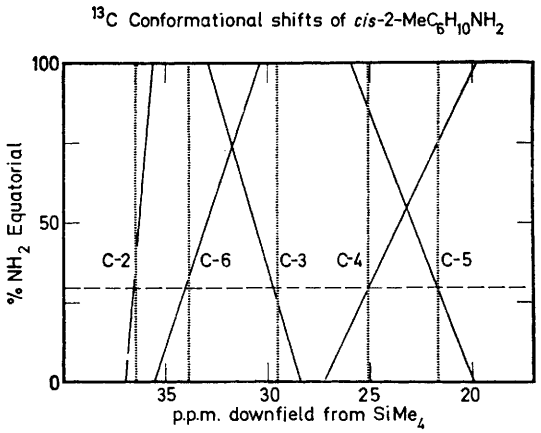
Compound	C-1	C-2	C-3	C-4	C-5	C-6	Me
(A) Conformationally homogeneous compounds in D_2O							
trans-2-Me-R [b]	56.64	40.64	34.94	26.74	26.31	(35.70) [e]	19.42
cis-3-Me-R	50.48	45.34	32.44	35.06	25.60	36.00	23.04
trans-4-Me-R	50.46	36.20	34.70	32.52	34.70	36.20	22.78
cis-,*cis*-3,5-Me_2R	50.20	44.76	31.88	44.04	31.88	44.76	22.86
trans-,*trans*-3,5-Me_2R	46.82	41.62	26.40	44.76	26.40	41.62	23.22
(B) Conformationally heterogeneous compounds in D_2O							
Cyclohexylamine	50.34	36.26	25.66	26.22	25.66	36.26	
cis-2-Me-R	51.60	(35.56) [e]	30.36	23.88	22.66	32.14	15.76
trans-3-Me-R	45.84	42.06	27.24	34.12	20.46	34.36	21.53
cis-4-Me-R	47.63	31.94	30.02	30.60	30.02	31.94	20.86
(C) Conformationally homogeneous compounds in C_6H_{12}							
trans-2-Me-R	57.24	41.52	34.92	(26.86) [d]	(26.49) [d]	37.40	19.34
cis-3-Me-R	51.18	46.72	32.52	(34.90) [e]	25.66	37.34	22.76
trans-4-Me-R	51.12	37.54	34.80	32.72	34.80	37.54	22.44
cis-,*cis*-3,5-Me_2R	50.82	46.06	32.00	44.10	32.00	46.06	22.60
trans-,*trans*-3,5-Me_2R	47.16	42.54	26.22	45.10	26.22	42.54	22.60
(D) Conformationally heterogeneous compounds in C_6H_{12}							
Cyclohexylamine	50.88	37.52	25.62	26.46	25.62	26.46	
cis-2-Me-R	51.58	36.44	29.54	25.06	21.70	33.84	16.84
trans-3-Me-R	46.34	43.10	26.84	(35.10) [e]	20.46	34.90	21.86
cis-4-Me-R	47.44	33.28	29.78	31.64	29.78	33.28	21.36

[a] p.p.m. downfield from $SiMe_4$ ± 0.02 p.p.m. [b] R = 1-$NH_2C_6H_{10}$. [c,d,e] Alternative assignments.

[11] M. W. Duch, Ph.D. Thesis, University of Utah, 1970.

[12] J. G. Batchelor, J. Feeney, and G. C. K. Roberts, *J. Magnetic Resonance*, 1975, **20**, 19.

Substituent Shift Parameters.—Although data were obtained for only a limited number of compounds it was possible to obtain a set of α, β, γ, and δ substituent parameters which predict the shifts of the conformationally homogeneous molecules to within *ca.* ± 0.2 p.p.m. (Table 2). The pattern of these shifts resembles those obtained by Dalling and Grant [10] for methyl substitution both in terms of the dependence on position and in terms of the dependence on conformation. The α substituent shifts due to amino-groups are larger than those due to methyl substitution because of the greater electronegativity of the amino-group and the γ_{eq}, δ_{eq}, and δ_{ax} shifts due to amino-groups are significant due to electric-field shift contributions.



^{13}C Chemical shifts of the ring carbon resonances of 2-*cis*-methylcyclohexylamine in C_6H_{12}. (——) predicted shifts as a function of conformation. (· · · ·) observed shifts. (— — —) calculated conformation

TABLE 2

Carbon-13 chemical shift parameters [a] indicating the effects of amino-substitution of a cyclohexane ring

Solvent	Substituent conformation	α	β	γ	δ
D_2O	Equatorial [b]	23.61	9.25	−1.18	−0.94
	Axial [c]	20.06	5.94	−6.73	−0.24
C_6H_{12}	Equatorial [b]	24.19	10.52	−1.16	−0.89
	Axial [c]	20.36	6.83	−6.95	0.05

[a] In p.p.m. downfield. [b] Three to five values available for each parameter. Range of values varies from ±0.09 to ±0.22 p.p.m. [c] Only one value available for each parameter.

It has not been possible to derive all the correction parameters required to predict the shifts when di-substitution occurs at adjacent or identical carbons. However, it does appear that the value of the parameter $\beta_e\gamma_e$ derived by Dalling and Grant [10] applies equally to 1-amino, 2-methyl substitution as to 1,2-dimethyl substitution and the average value derived for the $\alpha_e\beta_e$ parameter, −2.51 p.p.m. in D_2O solution and −2.73 p.p.m. in cyclohexane solution, is very similar to that found for dimethyl substitution, −2.45 p.p.m. For asymmetric substitution this single parameter may be useful for assignment purposes but is not sufficiently accurate in predicting the shifts of the substituted carbon resonances to enable these shifts to be used in the conformational analysis.

The shift of a γ methyl group due to a *gauche* amino-group is −3.60 p.p.m. in D_2O (−3.74 p.p.m. in cyclohexane) and follows the pattern that shifts of methyls due to γ *gauche* substituents are *ca.* 3 p.p.m. less than those of ring carbons.[10] In D_2O the substituent shifts of methyls at δ and ε positions were all < 0.25 p.p.m. In cyclohexane they were larger; −0.43 and −0.53 p.p.m. for δ and ε methyls due to an equatorial amino-group and −0.56 p.p.m. for a δ methyl due to an axial amino-group.

Significant differences exist between the substituent shifts derived for α, β, and methyl carbons in D_2O and in cyclohexane solutions. These differences are large enough that their neglect could occasionally lead to errors in assignment. The largest is for the β carbons, *ca.* 1.3 p.p.m.; and is clearly far too large to be ignored in estimating conformational equilibria.

Protonation Shifts.—The shifts observed upon protonation of the amino-groups of each compound are presented in Table 3. In general they follow the trends for acylic amines,[12] *i.e.* the protonation shift for a methine α carbon is downfield, that for a β methylene is large and upfield (*ca.* −5 p.p.m.) and those for γ and δ carbons are smaller, but again upfield. Similarly methyl substitution of a carbon leads to a less upfield shift, as recently predicted and observed in acyclic amines.[12] Consider, for example, C-2 in *trans*-2-methylcyclohexylamine, C-3 in *cis*-3-methylcyclohexylamine, and C-4 in *trans*-4-methylcyclohexylamine.

TABLE 3

Carbon-13 protonation shifts [a] of cyclohexylamine derivatives

Compound	Resonances C-1	C 2	C-6	C-3	C-5	C-4	Me
(A) Conformationally homogeneous compounds							
trans-2-Me-R [b]	+0.56	−4.06	−(4.30) [c]	−0.86	−1.22	−1.20	−0.78
cis-3-Me-R	+0.60	−5.98	−5.26	−0.78	−1.10	−1.20	−0.60
trans-4-Me-R	+0.62	−5.16	−5.16	−1.46	−1.46	−0.92	−0.62
cis-,*cis*-3,5-Me_2R	+0.62	−5.88	−5.88	−0.72	−0.72	−1.22	−0.62
trans-,*trans*-3,5-Me_2R	+2.28	−5.16	−5.16	+0.06	+0.06	−1.58	−0.78
(B) Conformationally heterogeneous compounds							
Cyclohexylamine	+0.78	−5.16	−5.16	−1.00	−1.00	−1.08	
cis-2-Me-R	+2.00	−(3.65) [c]	−5.18	+0.02	+0.32	−2.28	−1.66
trans-3-Me-R	+2.14	−5.36	−4.58	−0.20	−0.74	−1.78	−1.32
cis-4-Me-R	+2.04	−5.02	−5.02	−0.80	−0.80	−1.68	−1.02

[a] In p.p.m.; upfield shifts upon protonation negative, ±0.04 p.p.m. [b] R = 1-NH_2-C_6H_{10}. [c] Assignments may be reversed.

However, a very significant feature of the protonation shifts in Table 3 is the dependence on the number of *next nearest neighbours* of the observed carbon. The effect of a methyl substituent two bonds further than the observed carbon from the amino-group a ' β series ' effect, is to make

the protonation shift *more* upfield; *e.g.* compare C-2 (−5.98 p.p.m.) with C-6 (−5.26 p.p.m.) in *cis*-3-methylcyclohexylamine, and C-3 (−1.46 p.p.m.) in *trans*-4-methylcyclohexylamine with C-3 (−0.99 p.p.m.) in cyclohexylamine. The effect of a methyl substituent two bonds

NH_2–C–^{13}C–C–Me

(I)

β ' series ' methyl

from the observed carbon but the same number of bonds as the observed carbon from the amino-group, a ' β parallel '

Me

|

NH_2–C–^{13}C

(II)

β ' parallel ' methyl

effect, is to make the protonation shift *less* upfield, *e.g.* compare C-3 (−0.86 p.p.m.) with C-5 (−1.22 p.p.m.) in *trans*-2-methylcyclohexylamine.

Next nearest-neighbour contributions cannot be accounted for in terms of the simple additive scheme generally used for predicting ^{13}C chemical shifts. Fortunately they do not contribute to NH_2 substituent shifts but only to NH_3^+ substituent shifts. The prediction of the protonation shifts of a methylcyclohexylamine involves several sets of parameters derived from the data in Table 3, *i.e.* the protonation shifts which would be observed for the resonances of cyclohexylamine in the equatorial and axial conformations, the effects of immediate (α) substitution and the effects of nearest neighbour (β) substitution at each position (Table 4).

TABLE 4

Parameters describing protonation shifts [a] of cyclohexylamine derivatives

	Protonation shifts of cyclohexylamine ring		Methyl substitution effects [b]		
Resonance	Equatorial	Axial	α	β series	β parallel
Ring carbons					
α	0.60	2.28	*c*	−0.06	
β	−5.20	−4.44	1.14	−0.74	*c*
γ	−1.16	−0.34	0.40	−0.32	0.30
δ	−1.20	−1.58	0.30		0.00
Methyl carbons					
γ	−0.78	*c*			
δ	−0.62	−0.78			
ε	−0.62	*c*			

[a] Upfield shifts on protonation negative. In general only single observation available for each parameter. [b] Determined for diequatorial substitution. [c] Parameters not determined.

The derived protonation shift parameters in Table 4 show that there is considerable conformational dependence of the protonation shifts. Most surprising is the 1.68 p.p.m. difference between the shifts of an α carbon due to protonation of axial and equatorial amino-groups. Since this large difference arises only from the orientation of the γ and δ ring carbons relative to the amino-group it may be caused by a conformational dependence of the next nearest neighbour contributions to protonation shifts. The dependence of the protonation shifts of β carbons on conformation has been predicted to result from the orientation dependence of uniform-field linear electric-field shifts,[13] the observed direction of the conformational dependence being that predicted although its magnitude is somewhat smaller than expected. For γ and δ ring carbons a dependence of protonation shifts on conformation is expected as a simple result of the different distances between the amino-group and observed carbon in the axial and equatorial conformers.

Conformational Analysis.—The criterion used to determine the most probable conformation represented by the data for a given molecule is that the sum of squares of the deviations between observed and predicted shifts, weighted according to the conformational shift difference for each resonance, should be minimised. This condition is satisfied when

$$f_B = \sum_i \frac{(a_i - b_i).|a_i - b_i|.(a_i - x_i)}{\sum_i |a_i - b_i|^3} \qquad (1)$$

where f_B is the fraction of conformer B (NH_2 axial), a_i is the chemical shift of resonance i in conformer A (NH_2 equatorial), b_i is the chemical shift of resonance i in conformer B, x_i is the observed shift of resonance i, and the summation is over all non-equivalent resonances included in the analysis.

The uncertainty in the value of f_B was estimated from the variation of the conformers predicted on the basis of each individual resonance according to the formula

$$f_B = \frac{a_i - x_i}{a_i - b_i} \qquad (2)$$

Since the conformational shift differences may be either upfield or downfield for resonances within the same molecule, the differences between the conformations estimated from individual chemical shifts will tend to reflect random errors rather than systematic ones. They may therefore be used to estimate standard errors in the normal way.

(a) NH_2 *Derivatives.*—The chemical shifts of the ^{13}C resonances of the compounds with heterogeneous conformation (cyclohexylamine and *cis*-2-methyl-, *trans*-3-methyl-, and *cis*-4-methyl-cyclohexylamine) were calculated from the substituent shifts in Table 2 and ref. 10 for the conformers with equatorial (A) and axial (B) amino-groups. Because the accuracy with which the shifts may be predicted is *ca.* ± 0.2 p.p.m., those resonances for which the conformational shift difference was less than 1.5 p.p.m. were ignored. Similarly, all resonances for which the parameters necessary to predict their shifts are unknown (*i.e.* C-1, C-2, and C-Me in *cis*-2-methylcyclohexylamine) were omitted from the analysis.

The conformations derived from the remaining observed shifts and the shifts predicted for the individual conformers are presented in Table 5, together with the corresponding free-energy differences and the estimated errors of the derived values. The predicted shifts for the most probable conformations differ from the observed shifts by an average deviation per resonance ranging from 0.08 p.p.m. for cyclohexylamine in D_2O to 0.30 p.p.m. for *cis*-4-methylcyclohexylamine in D_2O. The corresponding standard errors of the estimated populations range from 2 to 12%, the most accurate determination of ΔG being for *cis*-2-methylcyclohexylamine, −2.2 ± 0.4 kJ mol^{-1} in cyclohexane and −0.6 ± 0.2 kJ mol^{-1} in D_2O.

(b) NH_3^+ *Derivatives.*—Although protonation shifts show

[13] J. G. Batchelor, *J. Amer. Chem. Soc.*, 1975, **97**, 3410.

a conformational dependence, they cannot be used alone to predict the conformations of compounds other than those which have very large conformational free-energy differences. This is because protonation changes the conformational free-energy difference and hence the conformation. Thus, such a protonation induced conformational change itself contributes to the observed protonation shift. For example, the shift of C_α in *cis*-4-methylcyclohexylamine due to protonation, +2.04 p.p.m., would indicate that the amino-group is 14% equatorial, whereas the population of

TABLE 5

Conformations and conformational free-energy differences of cyclohexylamine derivatives

Solvent	Compound	f_E [a]	ΔG/ kJ mol^{-1} [b]
R-NH_2 in C_6H_{12}	Cyclohexylamine (H-R) [c]	0.06 ± 0.05 [d]	6.9 ± 2.4 [d]
	cis-2-Me-R	0.70 ± 0.03	−2.2 ± 0.4
	trans-3-Me-R	0.74 ± 0.09	−2.6 ± 1.2
	cis-4-Me-R	0.77 ± 0.10	−3.1 ± 1.2
R-NH_2 in D_2O	Cyclohexylamine (H-R) [c]	0.06 ± 0.03	7.1 ± 1.3
	cis-2-Me-R	0.56 ± 0.02	−0.6 ± 0.2
	trans-3-Me-R	0.60 ± 0.06	−1.0 ± 0.7
	cis-4-Me-R	0.61 ± 0.12	−1.1 ± 1.3
R-NH_3^+ in D_2O	Cyclohexylamine (H-R [c])	0.03 ± 0.05	9.0 ± 3.8
	cis-2-Me-R	0.44 ± 0.02	0.6 ± 0.2
	trans-3-Me-R	0.56 ±0.09	−0.7 ± 0.9
	cis-4-Me-R	0.48 ± 0.13	+0.2 ± 1.3

[a] Fraction of conformer with axial NH_2. [b] For positive values, equatorial NH_2 is favoured. [c] R = 1-NH_2-C_6H_{10}. [d] Estimated standard error.

the conformer with an equatorial NH_2 group has been shown to be 39 ± 12% (Table 5). The discrepancy is due to an increase, upon protonation, in the population of the conformer having an equatorial amino-group.

However, it is possible to derive the conformations of the R–NH_3^+ species by predicting the shifts of each resonance for the axial and equatorial conformers and applying the same conformational analysis described above for the R–NH_2 species. In order to estimate the chemical shifts of the compound R–NH_3^+ it is necessary first to derive the chemical shifts of R–H using the parameters of ref. 9, then to add the appropriate NH_2 substituent shift parameters from Table 2 and finally to add the appropriate protonation shifts derived from Table 4. These protonation shifts cannot yet be predicted as accurately as the NH_2 substituent shifts because the next nearest neighbour contributions to protonation shifts are only known for the case where both methyl and amino-substituents are equatorial. At present these values must be assumed also to apply when one substituent is axial. However, the calculated conformational equilibria for R–NH_3^+ species, presented in Table 5 appear almost as reliable as those for R–NH_2, the most accurate determination of ΔG again being for *cis*-2-methylcyclohexylamine, a value of 0.6 ± 0.2 kJ mol^{-1}.

* It has been suggested that in 1,2-disubstituted cyclohexanes the conformational energies of the substituents will not be additive because the steric interaction between eq-ax *gauche* groups is greater than that between eq-eq *gauche* groups.[15] Chemical equilibration experiments, which measure the *configurational* energy difference between *cis*- and *trans*-isomers, do detect larger energy differences for 1,2-isomers than for 1,4-isomers. However, in the present experiments true *conformational* energies are measured. In each conformer of 1,2-*cis*-methylcyclohexylamine there is an *eq–ax gauche* interaction between the substituents and so the observed conformation free-energy is expected to be equal to the difference between the conformational free-energies of the substituents in monosubstituted cyclohexanes.

DISCUSSION

The calculated conformations in Table 5 indicate a trend towards a greater proportion of amino-groups becoming equatorial as the solvent is changed from aprotic to protic and as the amino-group is protonated.

In Table 6 is presented a comparison of the best data from the present study (for *cis*-2-methylcyclohexylamine) with a compilation of literature values [14] assuming that the conformational free-energies of the methyl

TABLE 6

Best values for conformational free-energy differences of methylcyclohexylamines [a]

Solvent conditions	ΔG(^{13}C n.m.r.) [b] / kJ mol^{-1}	ΔG(literature) [c] / kJ mol^{-1}
R-NH_2 in C_6H_{12}	−2.2 ± 0.4	−2.1 ± 0.8
R-NH_2 in D_2O	−0.6 ± 0.2	−0.4 ± 0.8
R-NH_3^+ in D_2O	0.6 ± 0.2	0.8 ± 0.8

[a] Those in which one substituent is axial and one equatorial. [b] Values for 2-methylcyclohexylamine from Table 5. [c] From ref. 14. $\Delta G(NH_2) - \Delta G(Me)$ assuming additivity (see footnote on this page) and using quoted best values: $\Delta G(Me)$ = 7.1 kJ mol^{-1}, $\Delta G(NH_2)$ = 5.0 (aprotic solvent), $\Delta G(NH_2)$ = 6.7 (protic solvent) and $\Delta G(NH_3^+)$ = 8.0.

and amino-groups are additive.* Under all solvent conditions there is good agreement within the specified limits of error. This comparison suggests that the high accuracy (± 0.2 kJ mol^{-1}) estimated for the conformational free-energy of *cis*-2-methylcyclohexylamine is, indeed, realistic. We can thus derive from the figures in Table 5 the following values for the changes in conformational free-energy differences arising from changes in solvent conditions:

$$\Delta G(NH_2)_{D_2O} - \Delta G(NH_2)_{cyclohexane} = 1.6 \pm 0.6 \text{ kJ mol}^{-1}$$

$$\Delta G(NH_3^+)_{D_2O} - \Delta G(NH_2)_{D_2O} = 1.2 \pm 0.4 \text{ kJ mol}^{-1}$$

These small changes in free-energy can be determined within ± 25%.

The estimated errors in determination of ΔG for the other compounds are rather larger. For the methylcyclohexylamines a major contributing factor is the relative size for each compound of the mean absolute conformational shift for the resonances included in the conformational analysis. In cyclohexane solution these are 5.46, 3.80, and 3.74 p.p.m. for *cis*-2-, *trans*-3-, and *cis*-4-methylcyclohexylamines. Since the accuracy of both $a_i - b_i$ and $a_i - x_i$ in equation (1) increases with the conformational-shift difference, it is not surprising that the results for *cis*-2-methylcyclohexylamine are more accurate than those of the other methylcyclohexylamines. The stated errors appear to be realistic because,

[14] J. A. Hirsch in 'Topics in Stereochemistry,' eds. N. L. Allinger and E. L. Eliel, Interscience, New York 1967, vol. 1, p.199.

[15] E. L. Eliel, S. H. Schroeter, T. J. Brett, F. J. Biros, and J. Richer, *J. Amer. Chem. Soc.*, 1966, **88**, 3327.

for any given solvent conditions, all the derived conformations and free-energy differences of the methylcyclohexylamines are consistent with each other and with the values predicted from previous work within the specified limits. However, the derived changes in conformational free-energy due to solvent change for *trans*-3- and *cis*-4-methylcyclohexylamine appear to be more accurate than might be expected from the large stated errors. This suggests that the errors involved in predicting the shieldings are not very solvent dependent so even for these compounds useful information could be obtained about the solvent dependence of conformation. For cyclohexylamine the free-energy difference is not measured accurately because it is so large (*ca.* 7.0 kJ mol^{-1}) that its dependence on the conformation, which is the variable actually measured, becomes very small. However, the conformation of cyclohexylamine can be measured quite accurately, within $\pm$ 5%.

Thus it appears that ^{13}C chemical shifts can now be used to determine the conformations of cyclohexane derivatives with an accuracy as good as $\pm$ 2%, although the level of accuracy depends on the size of the conformational-shift difference and so is lower for many compounds. The present success of the method should encourage further work to improve the prediction of chemical shifts and thereby approach more closely the very high potential accuracy of this method for measuring conformations.

[6/356 *Received*, 19*th February*, 1976]