Simulation of the Interconversion Path between Stable Conformations of the Furanose Ring: Methyl β-D-2-Deoxyribofuranoside and Simpler Ribose and Deoxyribose Analogues

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The results of simulation of the pseudorotation path in methyl β-D-2-deoxyribofuranoside, cyclopentane, tetrahydrofuran, 3-hydroxytetrahydrofuran, and 2,3-dihydroxytetrahydrofuran by using a geometrical ring-puckering model and an empirical all-atom force-field method are presented. For all these molecules energetic and geometric profiles of the *N–S* interconversion pathway are presented for various exocyclic group orientations. The difference in the simulated conformational behaviour of certain model compounds in the vapour state and in polar-solution conditions is discussed. The influence of the orientation and presence of exocyclic groups and the role of *C*- and *O*-ring structure on the conformational behaviour of the compounds examined is investigated. Conformational analysis of the furanose ring in several model compounds of increasing structure complexity reveals great diversity in their conformational behaviour in both geometric and energetic respects. The only features of conformational behaviour attributed to ring structure are: (*a*) the existence of energy minima in wide *N* and *S* regions of pseudorotation wheel; (*b*) the existence of *N–S* interconversion path *via* puckered-ring conformations. The puckering amplitude *q* varies considerably during *N–S* interconversion path and is no longer a quantitative feature of furanose-ring structure.

Conformational changes in the furanose ring are of great importance for the secondary structure of polynucleotide chains. The substantial differences between the A, B, and Z forms of a DNA double helix are correlated with the different furanose ring puckers.[1a]

The local changes of ring puckering in a polynucleotide chain lead to local perturbations of its secondary structure. The result of such a perturbation may be observed for example in the t-RNA molecule where $^3E \longrightarrow {}^2E$ puckering changes occur at intercalation sites or at positions where chain foldings switch abruptly from helical to looped.[1b]

Energy differences between the equilibrium *N* and *S* states in the furanose ring as well as the energy profiles of the *N–S* interconversion path are fundamental for conformational analysis of this system. Both *X*-ray crystallographic[2–5] and n.m.r. studies[6–9] suggest that the lowest energy *N–S* interconversion path occurs *via* pseudorotation rather than through inversion *via* a planar ring structure. Unfortunately it is not possible to follow the pseudorotation path in a furanose ring experimentally as it proceeds through unstable states. Experimental data are not sufficient to complete a description of this process and this is the reason why theoretical studies are performed to simulate the pseudorotation path in a furanose ring.[10–17] The aim of these studies is (*a*) to predict stable and intermediate conformations of the pseudorotation path, which may be achieved by plotting ring-puckering amplitude *q vs.* pseudorotation phase angle *P*[18] (geometric profile of the pseudorotation path); it is known from geometrical pseudorotation models[18,19] that each pair of *q*,*P* values describes a unique conformation of the five-membered ring; (*b*) to describe the pseudorotation barrier shown by plotting the energy of these conformations *vs.* phase angle (energetic profile of the pseudorotation path).

The aim of this study is to simulate the interconversion path between stable conformations of the furanose ring, in the sense mentioned above, in several model compounds in the vapour state and in polar solution, in order to understand the role of the sugar ring and exocyclic groups in conformational changes of deoxyribonucleoside structure. For simplification, the nucleoside molecule which contains a bulky heterocyclic base was substituted by methyl β-D-2-deoxyribofuranoside (MDRF), a molecule with a smaller methoxy group at C-1′. Up to now there have been no theoretical studies dealing with the conformational behaviour of this molecule except for one in which its stable conformations were simulated, reported by Wiorkiewicz-Kuczera and Rabczenko.[20] The comparison of the conformational behaviour of the MDRF molecule with data for nucleosides would answer the question: what is the role of particular nucleoside fragments in conformational behaviour. In order to determine the influence of exocyclic hydroxy groups on the conformational behaviour of ribose and deoxyribose analogues we also examined some simpler compounds, namely cyclopentane, tetrahydrofuran, 3-hydroxytetrahydrofuran (HT), and 2,3-dihydroxytetrahydrofuran (DHT). All model compounds examined in this work are presented in Figure 1.

The aim outlined in the previous section was achieved by minimizing the potential-energy function with respect to conformational degrees of freedom, without any change of phase angle *P*. Repetition of such a procedure for several *P* values yielded plots of optimized ring conformations (*q*), and their energies *vs.* phase angle *P*.

Method

In this study the nomenclature recommended by the IUPAC–IUB Joint Commission[21] has been applied. In accordance with this nomenclature, the phase angle $P = \varphi + 90°$ is used instead of the phase angle φ given in our previous paper.[18] $P = 0°$ and

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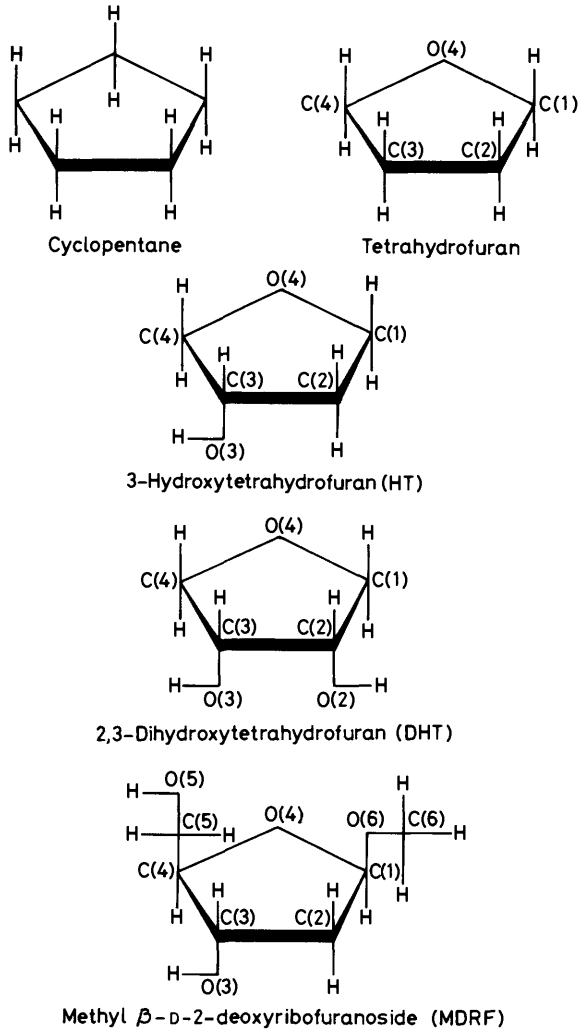

Figure 1. Model compounds examined in this study

$P = 90°$ involve the ring conformations ${}^{3}_{2}T$ and ${}^{5}E$, respectively. It should be noted that our phase angle, P, is not quite the same as the phase angle, P, applied in the (τ_m, P) pseudorotation formalism of Altona and Sungaralingam;[2] however, the difference is negligible. The empirical relationship between these two quantities was recently proposed by Harvey and Prabhakaran[22] in their molecular-dynamics simulation of the t-RNA^{Phe} molecule.

Molecular Geometry Construction.—Cartesian co-ordinates of ring atoms were calculated unequivocally for given q, P values by means of the geometric ring-puckering model HR.[18] The positions of hydrogen atoms were established in accordance with rules derived from the analysis of the neutron diffraction results of certain compounds by de Leeuv *et al.*[4] (*a*) Methylene hydrogen atoms show a tendency to retain local C_{2v} symmetry with the mean H–C–H angle equal to 107.6°: (*b*) Methine hydrogens on tertiary sp^2 carbon atoms are generally found in positions of equal bond angles to the other three carbon-atom substituents. Bond lengths and bond angles of the side chains were taken from appropriate experimental data. Cartesian co-ordinates of side chain atoms were calculated from internal co-ordinates mentioned above by means of the Thompson algorithm.[23]

Molecular-energy Calculations.—The Rasmussen potential-energy function[24,25] was adopted as the method of energy calculation. It was selected from several others[26–30] after test simulation of conformational behaviour of the tetrahydrofuran molecule using the geometric ring-puckering model HR.[18] The potential-energy function of a given molecule was minimized for several P values spread over the whole range, with variation of the puckering amplitude q and torsional angles of side chains. Only the conformational degrees of freedom were optimized, since full geometry optimization might have led, at best, to quantitative, and not qualitative differences in the energy profiles, and was too computationally time consuming. Minimization was effected by a simple iterative method with a constant step. The final steps were equal to 0.001 Å and 1° with respect to puckering amplitude and side-chain torsional angles, respectively.

Simulation of the Conformational Behaviour of Molecules in the Vapour State and in Polar Solution.—The lowest energy conformation obtained in the minimization energy procedure may be considered to be the most probable conformation of the isolated molecule. In order to characterize the conformational behaviour of the molecule in the vapour state as well as in polar solution, it is necessary to take into account all equilibrium conformations and pseudorotation paths between them. In this study of model compounds representing five-membered rings with side chains we introduced the mean geometric and energy profiles of the pseudorotation path in which the energy contributions corresponding to various conformers were incorporated with Boltzman weights. (*a*) In the case of the vapour state all possible rotamers were taken into account during the averaging procedure. (*b*) In the case of polar solution the number of rotamers was reduced on the assumption that in polar solution the molecules examined would prefer conformations which permit the formation of intermolecular hydrogen bonds. Thus exocyclic hydroxy groups should be orientated outside the molecule. This fundamental assumption of our simulation rejects rotamers involving OH • • • O(4) hydrogen interactions or those in which the exocyclic OH groups adopt a g^- conformation. Such an approach to the simulation of polar-solution conditions is based on experimental observations[31] and is also consistent with the analogy made by Wiorkiewicz-Kuczera and Rabczenko,[20,32] and by Lesyng and Saenger[33] in their theoretical studies. Such rules of simulation for solution conditions are very simple and do not include solute–solvent interactions in the calculations. However, it should be remembered that these interactions may be important for the modification of geometry of solute molecules.

Results

Cyclopentane.—The bond lengths were taken from an electron-diffraction study of the cyclopentane molecule,[34] b(C–C) = 1.546 Å, b(C–H) = 1.114 Å.

The energy difference between 'envelope' ($P = 18°$) and 'twist' ($P = 0°$) conformations is equal to $\Delta E = E_E - E_T = 0.09$ kJ mol^{-1}. The optimized q values are equal to 0.425 and 0.424 Å for 'twist' and 'envelope' conformations, respectively. These results are in accordance with the well known fact that free pseudorotation occurs in the cyclopentane molecule.

The optimized puckering-amplitude values may be compared with analogous values in the cyclopentane molecule derived from electron-diffraction data[34] only by means of the geometric ring-puckering model of Herzyk and Rabczenko (version for equilateral rings)[19] using the equation describing the dependence of mean non-bonded atom–atom distance R_{NB} on the puckering amplitude $Q[= (2/5)^{1/2}q]$ and ring bond length (Table 6 in ref. 19); R_{NB} and b(C–C) values are equal to

Table 1. Quantitative description of the energy profiles of the *N–S* interconversion pathway of HT and DHT molecules[a]

Rotamer	P_N	P_S	ΔE	$\Delta E^{E}_{N\rightarrow S}$	$\Delta E^{W}_{N\rightarrow S}$	$\Delta E^{E}_{S\rightarrow N}$	$\Delta E^{W}_{S\rightarrow N}$
HT molecule							
g^+	−2	165	−3.5	5.0	7.9	1.5	4.5
t	5	169	−3.7	3.9	7.6	0.3	4.0
g^-		200					
mean[b]	5	200	3.6	3.8	5.1	7.4	8.7
mean[c]	5	169	−3.6	4.1	7.6	0.5	4.0
DHT molecule							
tg^+	13	167	0.0	10.3	14.6	10.3	14.6
g^+t	11	169	0.0	3.9	12.4	3.9	12.4
g^-g^-	−18	198	0.0	43.0	3.1	43.0	3.1
g^+g^+	6	169	2.4	4.2	9.4	6.6	11.8
tt	11	174	−2.4	6.6	11.8	4.2	9.4
g^+g^-	4	193	14.8	3.8	1.4	17.6	16.1
g^-t	−13	176	−14.8	17.6	16.1	3.8	1.4
g^-g^+	−21						
tg^-		201					
mean[b]	−21	201	0.0	14.8	10.8	14.8	10.8
mean[c]	11	169	0.0	4.8	10.8	4.8	10.8

[a] P_N, P_S = phase angle of stable conformation in the *N* and *S* regions respectively (°); $\Delta E = E_N - E_S$ energy difference between the *N* and *S* local energy minima (kJ mol^{-1}); $\Delta E^{E}_{N\rightarrow S}$, $\Delta E^{W}_{N\rightarrow S}$ heights of energy barriers of $N \longrightarrow S$ interconversion path *via* east and west quadrant of pseudorotation wheel (kJ mol^{-1}). [b] Vapour state. [c] Polar solution.

2.444 ± 0.0015 and 1.546 ± 0.0015 Å, respectively,[34] the calculated *q* value being equal to 0.416 ± 0.012 Å. The same *q* value was obtained by Adams *et al.*[34] based on the same experimental data. In conclusion, it should be noted that the optimized value is consistent (within experimental error) with the value obtained from electron-diffraction data.

Tetrahydrofuran.—The bond lengths were taken from an electron-diffraction study of the tetrahydrofuran molecule,[35] *b*(C–C) = 1.536, *b*(C–O) = 1.428, *b*(C–H) = 1.115 Å.

The energy difference between the 'envelope' conformation (^5E, P = 90° in the *E* range or $_5E$, P = 270° in the *W* range) and the 'twist' conformation (^3_2T, P = 0° in *N* range or ^2_3T, P = 180° in *S* range) is equal to $\Delta E = E_E - E_T$ = 3.7 kJ mol^{-1}. The optimized *q* values obtained by means of our method are 0.360 and 0.330 Å for ^3_2T and ^5E conformations, respectively.

The calculated ΔE value is higher than the experimental values but these latter are not unequivocal and vary from 0.4 to 2 kJ mol^{-1} depending on the experimental method.[36–38] The lowest ΔE values obtained from far infrared spectroscopy data[37] and microwave spectroscopy data[36] were derived by means of the four-fold energy barrier model.

ab-initio Studies[39,40] concerning the *N–S* interconversion path in the tetrahydrofuran molecule give different ΔE values depending on the method applied. A recently obtained value,[40] ΔE = 3.1 kJ mol^{-1}, is similar to that obtained using our method. Authors of that study[40] assume that the lack of bond-length optimization accounts for such a high energy barrier.

The experimental value of the puckering amplitude derived from far infrared spectroscopy data[38] is 0.44 Å while the analogous value derived from electron-diffraction data[35] is 0.38 Å. It should be noted, however, that both these values were obtained based on the free pseudorotation model.

The *ab-initio* studies mentioned above[39,40] give different *q* values depending on the method applied. The recently published *ab-initio* 6-31G* study[40] gives values 0.370 and 0.362 Å for ^3_2T and ^5E conformations, respectively. The different energy values and different puckering amplitude values for ^3_2T and ^5E conformations obtained in this work as well as in the above-mentioned *ab-initio* studies[39,40] show that pseudorotation in the tetrahydrofuran molecule is not quite free in either the geometric or the energetic sense, as was the case in the cyclopentane molecule.

3-Hydroxytetrahydrofuran (HT) and 2,3-Dihydroxytetrahydrofuran (DHT).—Substitution of the 3-OH group (HT) and the 2- and 3-OH groups (DHT) for hydrogen atoms at C-2 and C-3 of tetrahydrofuran leads to substantial changes in the conformational behaviour of the ring. There are several geometric and energy profiles of interconversion pathways between stable conformations, depending on the hydroxy-group orientations. For clarity of description we have not presented graphically the geometrical and energy profiles of *N–S* interconversion pathway for each rotamer. Instead we present brief characteristics of each energy profile in Table 1, and the mean geometrical and energy profiles for the vapour state and solution in Figures 2 and 3, respectively.

3-Hydroxytetrahydrofuran (HT). The bond lengths had standard values:[41,42] *b*(C–C) = 1.54, *b*(C–O) = 1.43, *b*(C–H) = 1.10, and *b*(O–H) = 0.96 Å. Hydroxy groups were substituted in such a way that the angles between the C–OH bond and both C–C ring bonds, as well as C–O–H angle were constrained to 109.47°. The molecular energy minimization was carried out with respect to puckering amplitude *q* and torsional angle χ_3 = θ[C-2, C-3, O-3, H(O-3)] for phase angle *P* varying between 0 and 360° in steps of 18°. All three possible orientations of hydroxy group $g^+(\chi_3 = 60°)$, $t(\chi_3 = 180°)$, $g^-(\chi_3 = 300°)$ were taken into account during the minimization procedure.

There are two energy minima in the *N* and *S* regions of the pseudorotation wheel in the case of g^+ and *t* rotamers. The minima for g^+ and *t* rotamers are described by similar pseudorotation co-ordinates and correspond to the ^3_2T conformation (*N*) and to the conformation close to ^2E (*S*). This fact suggests that the changes of hydroxy group orientation between g^+ and *t* do not affect the stable conformations.

In the case of the g^- rotamer there is only one very deep and wide energy minimum in the *S* region of pseudorotation wheel, corresponding to the $_3E$ conformation of the smaller (than in the case of g^+, *t* rotamers) puckering amplitude *q*. This is a global energy minimum and is caused by the electrostatic interaction between H(O-3) and O-4. In the *N* region, however, a local energy minimum has not been found because of strong Lennard–Jones repulsion between the hydrogen atom of 3-OH group and hydrogens atoms of methylene groups juxtaposed to C-3.

2,3-Dihydroxytetrahydrofuran (DHT).—The bond lengths and the method of OH-group substitution were as in the case of the HT molecule. The presence of 2-OH and 3-OH groups introduces symmetry, so that the *N* and *S* states are related by a mirror plane through O-4 bisecting the C(2)–C(3) bond (plane *A*). This symmetry simplifies calculations in the sense that only the phase angle in the range −90° to +90° need be considered. Thus the molecular energy minimization was carried out with respect to puckering amplitude, *q*, and torsional angles, χ_2 = θ[C-1, C-2, O-2, H(O-2)] and χ_3 = θ[C-2, C-3, O-3, H(O-3)], for seven *P* values: −90, −72, −36, 0, 36, 72, and 90°. All nine combinations of OH-group orientations were taken into account during the minimization procedure. Of the nine possible rotamers, three are symmetric with respect to plane *A*, namely g^+t, tg^+, g^-g^- (the two symbols given describe the orientation of 2-OH and 3-OH, respectively). The other rotamers form mutually symmetric pairs (with respect to plane *A*), namely, g^+g^+ and tt; g^+g^- and g^-t; g^-g^+ and tg^-.

All rotamers except g^-g^+, and the corresponding symmetrical

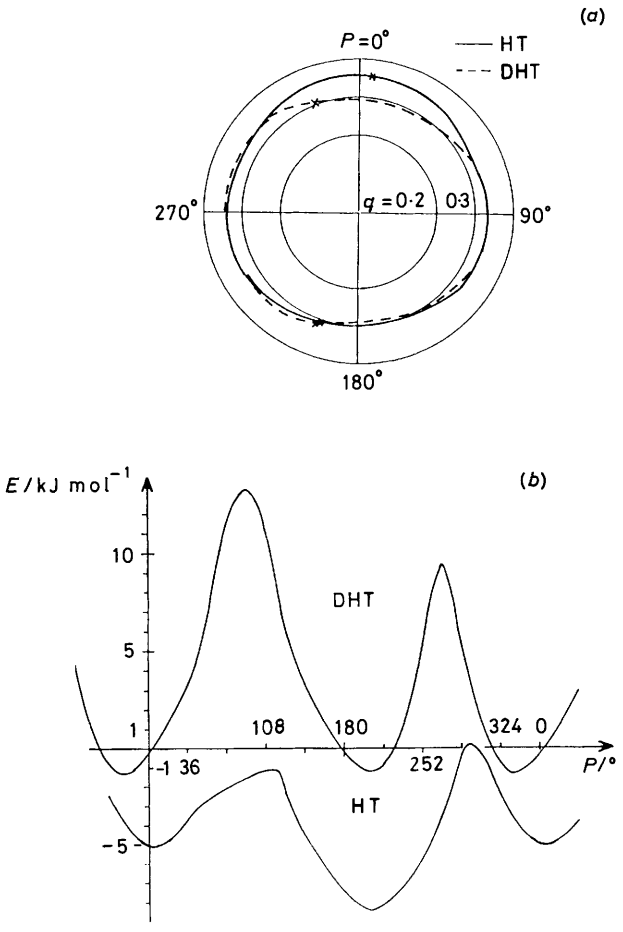

Figure 2. (*a*) Mean geometrical and (*b*) mean energy profiles of HT and DHT molecules in the vapour state

tg^-, display two energy minima in the *N* and *S* regions of the pseudorotation wheel (Table 1). All of these minima are described by phase angle *P* in the range -20 to $+15°$ in the *N* quadrant and between 165 and 200° in the *S* quadrant. The shapes and heights of energy barriers are different for different rotamers.

There is a large discrepancy between the conformational behaviour of rotamers in which at least one OH group adopts a g^- orientation and that of other rotamers. This situation is similar to that in the HT molecule: the transition of orientation of one OH group into g^- involves formation of an electrostatic interaction between the hydrogen atom of this group and ring oxygen atom O-4, and leads to the change of ring conformation from $^3T_2$ to $_2E$ in the *N* region or from $^2T_3$ to $_3E$ in the *S* region.

HT and DHT Molecules in the Vapour State.—There is a substantial difference between the conformational behaviour of HT and DHT molecules in vapour state (Figure 2, Table 1).

The mean energy profile of pseudorotation *T* pathway for the DHT molecule is symmetric with respect to the $P = 90°$ and $P = 270°$ axes. Both *N* and *S* minima lie in the west part of the pseudorotation wheel and correspond to $_2E$ and $_3E$ conformations, respectively. The west energy barrier is 10.8 kJ mol^{-1} and is *ca.* 4 kJ mol^{-1} lower than the east barrier. Thus, pseudorotation through the west part of the pseudorotation wheel is much more probable as the *N*–*S* interconversion pathway in the DHT molecule.

The mean geometric and energy profiles of the pseudorotation pathway in the HT molecule are asymmetric. The $_3E$ conformation in the *S* region is about 3.6 kJ mol^{-1} more stable than the ^3_2T one in the *N* region. The east energy barrier to the $S \longrightarrow N$ interconversion is 7.4 kJ mol^{-1} and is *ca.* 1.3 kJ mol^{-1} lower than the west. Thus, pseudorotation through the east part of the pseudorotation wheel is more probable as the *N*–*S* interconversion pathway in the HT molecule.

Based on the mean energy profile of the pseudorotation path the relative populations of *N* and *S* states were calculated for HT and DHT molecules from equations presented by Olson and Sussman.[43] In the case of molecules in the vapour state these populations are: HT, $\sigma_N = 0.23$, $\sigma_S = 0.77$; DHT, $\sigma_N = \sigma_S = 0.5$. The result obtained for DHT evidently arises because of the symmetry of the ring. The relative populations of HT may be compared with the corresponding ones obtained differently by Lesyng and Saenger[33] in their study concerning the prediction of stable conformations of HT and DHT molecules by means of the Rasmussen CFF method. These populations are: $\sigma_N = 0.36$, $\sigma_S = 0.64$. Because there are no experimental results on the *N*–*S* equilibrium of unsubstituted furanoses in the vapour state, it is impossible to compare theoretical results with measured data. However, the results obtained may be compared with those found by crystallographic structure analysis.[4] This last analysis gives $\sigma_N = \sigma_S = 0.5$ for ribonucleosides and $\sigma_N = 0.25$, $\sigma_S = 0.75$ for deoxyribonucleosides. Such a comparison should be treated only qualitatively, but nevertheless close agreement of results was achieved.

HT and DHT Molecules in Solution.—The difference in the conformational behaviour of HT and DHT molecules in polar solvents is not as pronounced as in the vapour state (Figure 3, Table 1).

In the case of the DHT molecule, the conformational *N*/*S* equilibrium remains unchanged because of the symmetry of the ring. The mean geometric and energy profiles (Figure 3) are symmetric with respect to the *W*–*E* axis of the pseudorotation wheel and $P = 90°$ or $P = 270°$ axes, respectively. Both *N* and *S* minima lie in the east part of the pseudorotation wheel, in contrast with the vapour state, and correspond to $^3T_2$ and $^2T_3$ conformations, respectively. The east energy barrier is 4.8 kJ mol^{-1} and is *ca.* 6 kJ mol^{-1} lower than the west one. Thus the *N*–*S* interconversion pathway through the east part of the pseudorotation wheel is much more probable than through the west part.

The mean geometric and energy profiles in the HT molecule are asymmetric. The ^3_2T conformation in the *N* region is *ca.* 3.6 kJ mol^{-1} more stable than the ^2E in the *S* region. The east energy barrier to the $N \longrightarrow S$ interconversion path is 4.1 kJ mol^{-1} and is *ca.* 3.5 kJ mol^{-1} lower than the west. Thus pseudorotation through the east part of the pseudorotation wheel is much more probable as the *N*–*S* interconversion pathway.

The relative populations of *N* and *S* states in HT and DHT molecules in solution are: HT, $\sigma_N = 0.74$, $\sigma_S = 0.26$; DHT, $\sigma_N = \sigma_S = 0.5$. As mentioned above, the result for the DHT molecule is evidently because of the symmetry of the ring. In the case of the HT molecule, however, the inversion of *N*/*S* relative populations is observed. A similar inversion ($\sigma_N = 0.84$, $\sigma_S = 0.16$) was obtained by Lesyng and Saenger in their study[33] mentioned in the previous section, where similar rules of solution conditions simulation were adopted.

Methyl β-D-2-*Deoxyribofuranoside* (*MDRF*).—*The MDRF molecule in solution.* The bond lengths and exocyclic bond angles of this compound are presented in Table 2. Only the conformational behaviour of this molecule in solution was simulated since experimental data dealing with preferential conformations of methyl β-furanosides in solution already exist.[44] Only the rotamers in which the 3-OH adopts g^+ and *t*

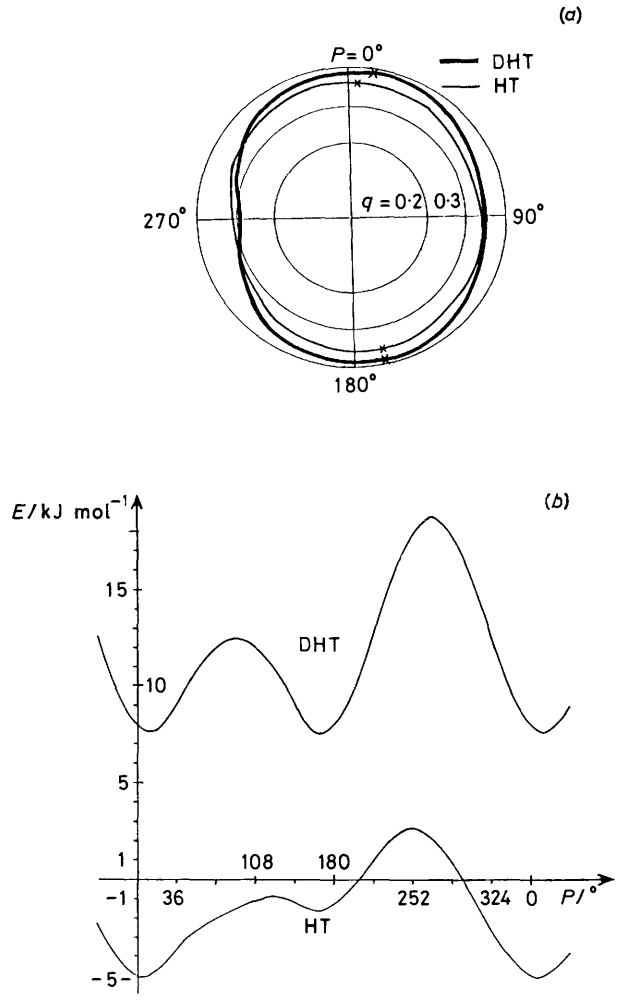

Figure 3. (*a*) Mean geometrical and (*b*) mean energy profiles of HT and DHT molecules in polar solution

Table 2. Bond lengths and exocyclic bond angles of the MDRF molecule

Bond lengths/Å		Bond angles/°	
C(1)–C(2)	1.529[a]	O(4)–C(1)–O(6)	106.0[b]
C(2)–C(3)	1.527[a]	C(2)–C(1)–O(6)	110.2[b]
C(3)–C(4)	1.526[a]	C(1)–O(6)–C(6)	114.3[b]
C(4)–O(4)	1.449[a]	C(2)–C(3)–O(3)	111.2[b]
O(4)–C(1)	1.411[a]	C(4)–C(3)–O(3)	111.9[b]
C(1)–O(6)	1.423[b]	C(3)–O(3)–H	108.4[b]
O(6)–C(6)	1.420[b]	C(3)–C(4)–C(5)	114.2[b]
C(4)–C(5)	1.525[b]	O(4)–C(4)–C(5)	108.3[b]
C(5)–O(5)	1.428[b]	C(4)–C(5)–O(5)	108.6[b]
C(3)–O(3)	1.424[b]	C(5)–O(5)–H	109.9[b]
C–H	1.10[c]	H–C(6)–H	109.47
O–H	0.96[c]		

[a] The ring bond lengths are the mean values of the set of 178 β-D-furanoside fragments known from crystallography.[4] [b] Mean values of the set of corresponding fragments optimized in different local energy minima (ref. 20 and personal communication). [c] Standard values (refs. 41 and 42).

orientations, and the 5-OH group the *t* orientation, were taken into consideration in simulating the solution conditions. The potential-energy function was minimized with respect to the puckering amplitude, *q*, and all exocyclic torsional angles for the phase angle *P* in the range 0—360° in steps of 36°. The following

Table 3. Quantitative description of the energy profiles of the *N*–*S* interconversion path of the MDRF molecule[a]

Rotamer	P_N	P_S	ΔE	$\Delta E^{E}_{N\rightarrow S}$	$\Delta E^{W}_{N\rightarrow S}$	$\Delta E^{E}_{S\rightarrow N}$	$\Delta E^{W}_{S\rightarrow N}$
anti							
$\gamma^- t$	−21	207	−6.6	8.1	8.8	1.5	2.2
$\gamma^- g^+$	−25	195	−4.4	8.4	6.5	4.0	2.1
$\gamma^t t$	−18	207	−7.1	9.6	10.5	2.5	3.4
$\gamma^t g^+$	−21	202	−8.4	11.1	10.6	2.7	2.2
$\gamma^+ t$	−7	194	−2.6	4.4	7.0	1.8	4.4
$\gamma^+ g^+$	−11	184	−3.6	7.0	7.7	3.4	4.1
high anti							
$\gamma^- t$	−24	234	−7.8	12.3	8.7	4.5	0.9
$\gamma^- g^+$	−26	241	−6.8	13.8	7.3	7.0	0.5
$\gamma^t t$	−24	225	−7.3	13.3	8.8	5.0	1.5
$\gamma^t g^+$	−21	230	−9.0	15.5	9.4	6.5	0.4
$\gamma^+ t$	−9	225	−7.2	10.4	9.0	3.2	1.8
$\gamma^+ g^+$	−15	216	−8.0	13.2	8.8	5.2	0.8

[a] Parameters as in Table 1.

possible exocyclic torsional angles values were established as initial values in the minimization procedure: χ_1 = θ(O-4, C-1, O-6, C-6) = 180° (*anti*); χ_1 = 300° (*high anti*); θ(C-1, O-6, C-6, H) = 60°; χ_3 = θ[C-2, C-3, O-3, H(O-3)] = 60° (g^+), χ_3 = 180° (*t*); χ_4 = θ(C-3, C-4, C-5, O-5) = 60° (γ^+), χ_4 = 180° (γ^t), χ_4 = 300° (γ^-); θ[C-4, C-5, O-5, H(O-5)] = 180°. Thus 12 rotamers were taken into account in the minimization procedure: $\gamma^+g^+, \gamma^+t, \gamma^tg^+, \gamma^tt, \gamma^-g^+, \gamma^-t$, for both *anti* and *high anti* conformations about the C(1)–O(6) bond.

There are several geometric and energy profiles of the interconversion path between stable conformations depending on side-chain orientations. For clarity of description, we have not presented them all. Brief characteristics of the energy profiles are presented in Table 3, and the mean geometric and energy profiles are presented in Figure 4.

All the rotamers examined display two energy minima each in the *N* and *S* regions; however, the *N* minimum is of lower energy in each case. All *N* local minima lie in the *P* range −30 to −6°. In the case of the *S* state, these *P* ranges are different for *anti* and *high anti* rotamers and are equal to 180—210° and 216—238°, respectively. As is known from a statistical analysis of crystallographic nucleoside and nucleotide fragments,[4] the phase angle ranges of stable conformations of 2′-deoxyribonucleosides and -nucleotides are *N*(−1°, +25°) and *S*(146°, 213°). The first range corresponds to ${}^3_2T$ and ${}^3E$ conformations while the broad *S* range is centred around the ${}^2E$ conformation. The discrepancy between the positions of stable conformations in the *N* and *S* regions of the pseudorotation wheel in the above-mentioned crystallographic structures and in the MDRF molecule is caused by the change in substitution at C-1. The small methoxy group can be oriented in the more axial position (${}_2E$ or ${}_3E$) which is not available for the large heterocyclic base owing to its repulsive interaction with the hydroxymethylene group at C-4.

All *high anti* energy profiles have essentially lower energy than the corresponding *anti* ones. This situation is at variance with that for nucleosides and nucleotides in which an *anti* orientation of the heterocyclic base is generally preferred. The *high anti* orientation of non-modified bases is not preferred because of steric hindrance between hydrogen atoms, namely pyrimidine 6-H or purine 8-H and sugar hydrogens.[1c] In the case of the small and non-flat methoxy group, such restrictions do not exist. In this case both *anti* and *high anti* conformations about the C(1)–O(6) bond are possible; however, the latter is more stable because of the existence of weak electrostatic interactions between methyl group hydrogen atoms and O-4.

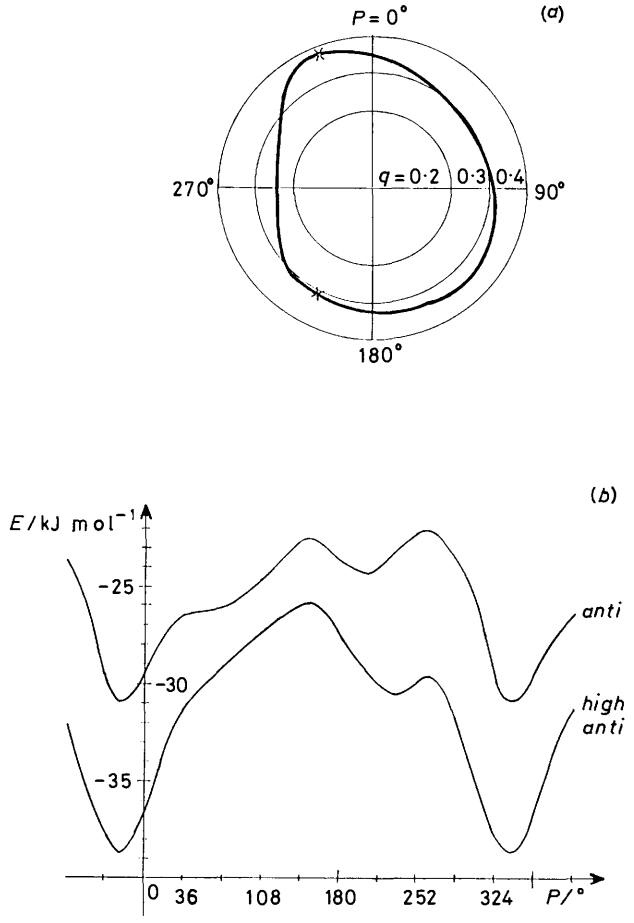

Figure 4. (*a*) Mean geometrical profile *anti* and (*b*) mean energy profiles (*anti*, *high anti*) of the *N*–*S* interconversion path of the MDRF molecule in polar solution

In contrast with the strong dependence of the shapes of the energy profiles of pseudorotation on the orientation of the methoxy group, an analogous dependence with respect to geometric profiles has not been found.

In the *N* region of the pseudorotation wheel, the order of preferred conformations about the C(4)–C(5) bond is as follows: γ^-, γ^t, γ^+, regardless of the orientation of the methoxy group. In other words, for a given orientation about the C(1)–O(6) bond the conformational energy in the *N* state depends on the orientation of hydroxymethylene group. In the *S* region a preferred conformation about C(4)–C(5) bond is γ^-; however, further order depends on the orientation at the χ_1 and χ_3 angles.

A correlation between the conformation about the C(4)–C(5) bond and the shape of the energy profile has not been found; however, such a correlation exists with respect to the geometrical profile of the *N*–*S* interconversion pathway. The γ^t geometrical profile seems to be rounded while the γ^+ profile is flat in the west part of the pseudorotation wheel. Thus in the case of the γ^+ rotamer, the *N*–*S* interconversion pathway through the west part of pseudorotation wheel leads to an extremely low value for the puckering amplitude *q*, regardless of the height of the energy barrier.

Two mean energy profiles have been obtained for *anti* and *high anti* orientations of the methoxy group (Figure 4). The mean (over the *anti* rotamers) energy profile of the *N*–*S* interconversion pathway of the MDRF molecule in solution displays two energy minima in the *N* and *S* regions of the pseudorotation wheel, west of the *N*–*S* axis (Figure 4). These *N* and *S* minima correspond to ${}_2E$ and ${}_3T^4$ conformations, respectively. The *N* minimum is the lower in energy of the two, $\Delta E = E_S - E_N = 6.7$ kJ mol^{-1}. Both energy barriers are comparable, the east barrier being about 0.4 kJ mol^{-1} lower than the west, which makes the east pseudorotation path slightly more probable than the opposite one. The lowest energy barriers to the *N*–*S* interconversion pathway are equal to $\Delta E^{E}_{N\rightarrow S} = 8.6$ kJ mol^{-1} and $\Delta E^{E}_{S\rightarrow N} = 1.9$ kJ mol^{-1}. In the case of *high anti* rotamers the mean energy profile of *N*–*S* interconversion path of MDRF molecule in solution displays, incremental energy differences between stable conformations ${}_2E$ in the *N* state and ${}^4E$ in the *S* state; $\Delta E = 8.1$ kJ mol^{-1}. The west energy barrier is *ca.* 3.7 kJ mol^{-1} lower than the east, which makes the west pseudorotation path the more probable of the two. The lowest energy barriers to *N*–*S* interconversion are $\Delta E^{W}_{N\rightarrow S} = 9.1$ kJ mol^{-1} and $\Delta E^{W}_{S\rightarrow N} = 1$ kJ mol^{-1}.

The relative populations of the *N* and *S* states in the MDRF molecule in solution are: $\sigma_N = 0.86$, $\sigma_S = 0.14$ for the *anti* rotamers and $\sigma_N = 0.94$, $\sigma_S = 0.06$ for the *high anti* rotamers.

The preference for *N*-type puckering in the MDRF molecule in solution is consistent with ^{1}H n.m.r. results of Gerlt and Youngblood[38] obtained for this compound and also with ^{13}C n.m.r. measurements of Cyr and Perlin[45] for methyl D-ribofuranoside. The theoretical studies of Wiorkiewicz-Kuczera and Rabczenko[20,32] also reveal the preference of *N*-type conformation in methyl β-D-furanosides in solution.

Discussion

Influence of Polar-solution Conditions on the Conformational Behaviour of Certain Model Compounds.—The 'transfer' of HT and DHT molecules from the vapour state to polar solution leads to essential differences in their conformational behaviour. From the mean energy profiles of the *N*–*S* interconversion pathway (Figures 2 and 3), it can be seen that such a transfer causes the change of the stable conformations ${}_2E \longrightarrow {}^3T_2$ (*N*) and ${}^3E \longrightarrow {}^2T_3$ (*S*) in the DHT molecule and ${}_3E \longrightarrow {}^2E$ (*S*) in the HT molecule. The ${}^3_2T$ *N*-type conformation of the HT molecule does not change. These conformational transitions occur owing to the lack of attractive electrostatic interaction between the OH group(s) and O-4 in solution, in contrast with the situation for isolated molecules.

The 'transfer' of both molecules to polar solution makes the east pseudorotation barrier essentially lower than the west. Furthermore, it makes the lowest energy barrier to pseudorotation lower than in the case of vapour state. In other words it is easier for a molecule in solution to change the type of puckering.

In the case of the asymmetric HT molecule such a transfer also causes complete inversion of the *N*/*S* population ratio from 23:77 in the vapour state to 74:26 in solution.

The MDRF molecule in aqueous solution also displays the preference for *N*-type puckering of the ring. However, in almost all nucleosides and mononucleotides in solution the sugar ring has been observed to prefer the ${}^2E$ conformation of the *S*-type.[31,46] Thus the presence of a heterocyclic base at C-1′ imposes the *S*-type ring conformation, in contrast with the unsubstituted sugars which prefer the *N*-type ring conformation in aqueous solution.

Influence of the Orientation of Exocyclic Groups on the Conformational Behaviour of the Furanose Ring.—The conformational behaviour of the furanose ring may be considered from both a geometric and an energetic point of view: (*a*) the geometric behaviour characterized by stable conformations and geometric profiles of interconversion, and (*b*) the energetic behaviour characterized by the energies of the stable conformations and the energy profiles of interconversion.

In the case of HT and DHT molecules in solution, the influence of the orientation of OH groups on the geometric and energetic behaviour of the furanose ring is negligible. However, if these compounds are in the vapour state and if g^- orientations of the OH groups are assumed, the influence of the orientation of the hydroxy group is apparently significant. The conformational behaviour of rotamers in which at least one OH group adopts the g^- orientation is significantly different from the other rotamers because of the existence of electrostatic attraction between the g^- OH group and the ring oxygen atom.

In the case of the MDRF molecule in solution both geometric and energetic behaviours of the furanose ring are affected by the orientations of exocyclic groups at C-1 and C-4. The orientation of the methoxy group at C-1 strongly influences the energies of stable conformations of the MDRF molecule and the energetic profile of the interconversion pathway between them. However, the methoxy group orientation is not important either for the geometries of stable conformers or for the geometric profile of the pseudorotation path. On the other hand, the orientation of the hydroxymethylene group at C-4 affects the geometric behaviour of the sugar ring but does not affect the energetic behaviour. The influence of hydroxy-group orientation at C-3 on the conformational behaviour of the MDRF molecule in solution seems to be negligible.

Since different factors influence the geometric and energetic behaviour of the sugar ring there is no correlation between the two as can be seen in Figure 4.

Conformational Behaviour of the Furanose Ring with Different Exocyclic Groups.—If the results presented in this study are examined, the following question arises: which of the features of the conformational behaviour of model compounds may be considered to be intrinsic properties of C- and O-ring structure, and which are caused by the presence of side chains?

Tetrahydrofuran may be regarded as a model of a furanose ring with no side chain. In this case the description of the conformational behaviour in terms of energetic and geometric profiles of the *N–S* interconversion path is simple. Both profiles are symmetric and related to each other. It could be said that the C- and O-ring structure has two stable, symmetric conformations ${}^3_2T$ and ${}^2_3T$ in the middle of *N* and *S* regions of the pseudorotation wheel, respectively, with two symmetric energy barriers in the middle of *E* and *W* quadrants. The geometric profile of the *N–S* interconversion path is qualitatively closer to the free pseudorotation line than to inversion *via* a planar conformation; however, the puckering amplitude, *q*, is dependent on the energy of a given conformation; *q* decreases with energy increase.

If we examine the mean energetic and geometric profiles of *N–S* interconversion path for HT, DHT, and MDRF molecules, some similarities and differences are found, as compared with the tetrahydrofuran molecule: (*i*) All energetic profiles have two energy minima in the *N* and *S* regions of the pseudorotation wheel centred at ${}^3_2T$ and ${}^2_3T$ conformations. Stable conformations of the ring inside the *N* and *S* regions of the pseudorotation wheel are different if compounds with different exocyclic groups are considered, *i.e.* if the DHT molecule in solution or 2′-deoxyribonucleosides[4] are compared with the MDRF molecule.

(*ii*) Energy differences between stable conformations and relative populations of *N* and *S* states are different for model compounds of different symmetry, as in the case of the HT and DHT molecules. The symmetry of the molecule with respect to the plane through O-4 bisecting the C(2)–C(3) bond (tetrahydrofuran and DHT molecules) involves the symmetries of both energetic and geometric profiles of the pseudorotation pathway with respect to the $P = 90°$ and $P = 270°$ axes and the *W–E* axes, respectively. In the case of asymmetric compounds (HT and MDRF) both profiles are asymmetric. We obtained a similar *N*/*S* population ratio for these molecules in polar solution conditions, which could indicate that this quantity is characteristic of asymmetric 2′-deoxy compounds in solution. However, n.m.r. data for 2′-deoxyribo-nucleosides and -nucleotides[31,46] show a reversed *N*/*S* population ratio.

(*iii*) The mutual dependence of the height of the energy barriers is significantly different for *anti* and *high anti* rotamers of the MDRF molecule. Moreover, the essentially higher west energy barrier in nucleosides and nucleotides[1] is not so in MDRF molecule (Figure 4) because of the smaller side group at C-1 in the latter molecule.

(*iv*) Geometric profiles of the *N–S* interconversion path are quantitatively different for different model compounds; however, they are always qualitatively closer to the free pseudorotation route than to inversion *via* a planar conformation. This observation led us to conclude that the free-pseudorotation-like, qualitative character of the geometric profile arises from the nature of the ring, but the presence and conformation of side groups deforms it seriously in a way difficult to predict. From a detailed analysis of sugar puckering in a large series of nucleosides it can be seen that the puckering amplitude shows a unimodal distribution.[1d,4] Because of this some authors state that the puckering amplitude is independent of the pseudorotation phase angle, and of the presence and orientation of side chains, and can therefore be regarded as an intrinsic propety of C- and O-ring structure.[1d,47] Our results reveal that in the furanose ring the puckering amplitude, *q*, is substantially dependent on the phase angle, *P*. This fact may be confirmed by some earlier studies[13,5] and by a recent molecular-dynamics simulation of the t-RNA^{Phe} molecule.[22] Qualitative mutual similarity of geometric profiles for different model compounds cannot justify the treatment of the puckering amplitude, *q*, as independent of *P*. This is true only for the cyclopentane molecule, and more for complicated compounds should be treated as a considerable approximation.

Taking into account all above-mentioned remarks we conclude that the only intrinsic properties of the furanose ring structure are: (*a*) the existence of energy minima in wide *N* and *S* regions of the pseudorotation wheel, and (*b*) the existence of *N–S* interconversion path *via* puckered conformations. Thus the geometric and energetic flexibilities of the furanose ring are described mainly by the presence and orientation of the side chains and therefore conformational behaviour of one model compound cannot be a pattern for the other with different exocyclic groups.

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