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INFLUENCE OF THE ORIENTATION OF HYDROXYL GROUPS ON THE PUCKERING MODES OF FURANOID RINGS

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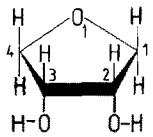
ABSTRACT

The influence of the orientations of HO-2 and HO-3 on the puckering of the ring in (2*S*,3*R*)-tetrahydrofuran-2,3-diol (**1**) and (3*R*)-tetrahydrofuran-3-ol (**2**) has been investigated by consistent force-field methods. In the E or W regions of the pseudorotation cycle, there are no local energy minima. The local minima in the N and S regions are affected by different orientations of the hydroxyl groups, with up to 12.2 kJ.mol^{-1} for **1** and 6.9 kJ.mol^{-1} for **2**. If statistical weights of different rotamers are used to calculate N/S equilibrium constants, both conformational states are equally populated. However, if the orientations of the hydroxyl groups are restricted to simulate behaviour of these molecular fragments in DNA double-helices, the equilibrium shifts towards N. The results indicate that, in the vapor state, a unique one-dimensional N–S energy-barrier does not exist and that there is a spectrum of barriers depending on the orientation of the hydroxyl groups.

INTRODUCTION

The stereochemistry of furanoses has been studied extensively[1–8]. The transition of puckering modes, now generally defined in terms of pseudorotation parameters P (phase angle) and τ_m (puckering amplitude), was found to occur essentially without an energy barrier[2], whereas more recent results point to a barrier



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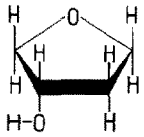

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of 7–15 kJ.mol^{-1} between the two main puckering modes C-2′-*endo* (south or S) and C-3′-*endo* (north or N)[6–8].

We have carried out a thermodynamic study employing a consistent force-field method with a new parametrisation not used previously. We have also

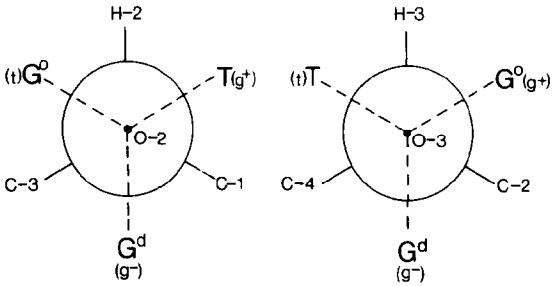

Fig. 1. The geometrical (G°,T,G^d) and the systematic (g$^+$,t,g$^-$) descriptions of orientations of the O–H groups; G°, *gauche* orientation of an O–H group with respect to the C-2–C-3 bond and pointing *outside*; T, *trans* orientation of an O–H group with respect to the C-2–C-3 bond; G^d, *gauche* orientation of an O–H group with respect to the C-2–C-3 bond and pointing *down* the ring; g$^+$, t, and g$^-$ denote the orientations given by the C-1–C-2–O-2–H and C-2–C-3–O-3–H angles for the HO-2 and HO-3 groups, respectively. The TG° (g$^+$g$^+$) configuration denotes the *trans* orientation of HO-2, and *gauche outside* for HO-3.

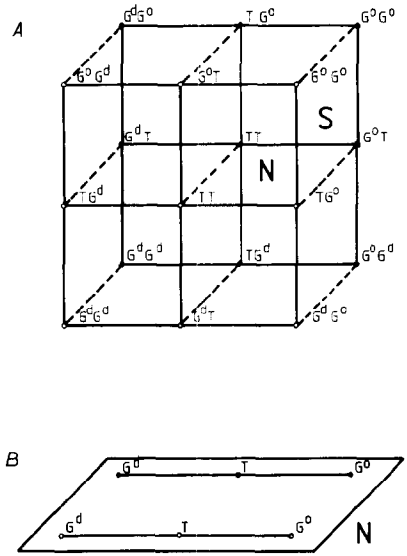

Fig. 2. *A*. Schematic representation of the possible conformational states of **1**. On the front (back) surface, the local minima of the energy, corresponding to all possible configurations of the O–H groups in the N (S) conformation of the ring, are presented. The two symbols given describe the orientation first of HO-2 and second of HO-3. Note that (*1*) the energy states do not differ if the orientation symbols in N/S are permutated; (*2*) in general, the states of the same orientation of the O-H groups (in the N and S conformation) possess different energies. Exceptions are the "diagonal" states G^{d}G^d, TT, and G°G°. *B*. Schematic representation of the possible conformational states of **2**. In general, all states possess different energies.

considered how different orientations of HO-2 and HO-3 influence the population of the puckering modes in simple model compounds, because an *ab initio* calculation for ethane-1,2-diol indicated that, in the preferred *gauche* form, the total energy varies in the range 0–26 kJ.mol^{-1} when the hydroxyl groups are in different orientations[9].

The model compounds selected were (2*S*,3*R*)-tetrahydrofuran-2,3-diol (**1**) and (3*R*)-tetrahydrofuran-3-ol (**2**), which are formally related to ribofuranose and 2-deoxyribofuranose. In **1**, the presence of HO-2,3 introduces symmetry, so that the E and W states of sugar puckering contain a mirror plane through O-1 and bisecting the C-2–C-3 bond, and the N and S states are related by this mirror symmetry. Therefore, simplifications in the calculations are possible, in contrast to those for the asymmetric molecule **2**.

Each hydroxyl group can adopt three different orientations with respect to the bond to the corresponding carbon. These orientations for **1** and **2** are displayed in Fig. 1. Because there are two hydroxyl groups in **1**, there are 3 × 3 possible conformations, and 3 for **2** with only one hydroxyl group. Multiplying these with the number of main puckering modes (N and S), 18 different conformers are obtained for **1** and 6 for **2**, which are shown schematically in Fig. 2.

METHODS

The statistical-mechanical method was used, assuming that each molecule is in thermodynamic equilibrium with its surroundings and that its partition function can be factorised[10–12]. The programme employed was the consistent force-field method (CFF) of Niketić and Rasmussen, which was developed to account simultaneously for equilibrium geometry, vibrational spectra, and thermodynamic data of a set of related compounds[13–17]. The force field used consisted of harmonic functions for the bond and angle deformations. Lennard-Jones potentials and Coulomb terms accounted for non-bonded interactions between atoms separated by three or more bonds.

All initial conformations of the rings of **1** and **2** were generated by the geometrical programme described in ref. 18. The conformations were obtained for $\tau_m = 39°$ and $0° \leq P \leq 360°$ with an increment of 10° or 20°. For each conformation of the rings of **1** and **2**, all possible orientations of the hydroxyl groups were probed. The initial conformations were minimised with respect to all degrees of freedom through 20–50 steepest descent iterations followed by 20–50 steps of a modified Newton algorithm and/or the Davidon–Fletcher–Powell minimisation method. Minimisation was finished when the quadratic norm of the gradient became $<10^{-6}$ kJ.mol^{-1}.Å^{-1}. The minima of the potential-energy surface were related to equilibrium conformations.

The normal vibration calculations and translation-rotational corrections allowed determination of the partition function and hence calculation of the enthalpy, entropy, and free enthalpy.

As derived by previous studies and also consistent with the present work, furanoses adopt two sets of conformational states, termed N or S according to their location on the pseudorotation cycle[19]. If preferences of one state over another are to be presented, relative populations are used which, in general, are given by the Boltzmann distribution:

$$\sigma_q = (1/Z) \exp(-V_q/RT). \tag{1}$$

In this equation, q = N and S, and V_N and V_S denote the energies of N and S conformational states, with

$$Z = \sum_q \exp(-V_q/RT). \tag{2}$$

In a more sophisticated, classical approach[7,8], the partition function is obtained by integration of the Boltzmann energy factors V over the whole pseudorotation cycle P, yielding

$$Z_p = \int_{P=0}^{2\pi} \exp[-V(\tau_m^\circ, P)/RT]\, dP, \tag{3}$$

and statistical weights can be expressed as

$$\sigma_p = (1/Z_p) \int_P \exp[-V(\tau_m^\circ, P)/RT] dP. \tag{4}$$

The integration over P assumes constant τ_m although, in fact, τ_m depends slightly on the phase angle P and therefore $V = V[\tau_m(P), P]$.

This method does not take into account different orientations of the hydroxyl group(s), which are considered here, and, because of its classical nature, neglects oscillation corrections.

Therefore, the calculated free-enthalpy differences between two conformational states 1 and 2 of a molecule,

$$\Delta G_{12}^\circ = \Delta H_{12}^\circ - T\Delta S_{12}^\circ, \tag{5}$$

are used here directly to derive, on the basis of equilibrium statistical thermodynamics, the relative population (the equilibrium constant) of the states expressed as:

$$K_{12} = \sigma_1/\sigma_2 = \exp(-\Delta G_{12}^\circ/RT). \tag{6}$$

The free-enthalpy differences ΔG°_{12} can be calculated for all possible conformers of the furanose ring. Because different orientations of the hydroxyl groups can be included, a more exact description of the conformational behaviour of the furanose ring becomes possible.

RESULTS AND DISCUSSION

Starting with the orientations of hydroxyl groups defined in Fig. 1 and varying the pseudorotation phase-angle P in steps of 10°, consistent force-field calculations were performed and the respective geometries allowed to optimise. During this latter process, the relative orientations of the hydroxyl groups changed only several degrees from their starting positions. An exception is the G^dG° configuration of the ring of **1** in S-puckering mode. This particular state is sterically unfavourable and therefore the hydroxyl groups have to rotate into more suitable positions. For all configurations of the hydroxyl groups investigated, the rings of **1** and **2** display two separated local energy-minima in the N and S puckering modes.

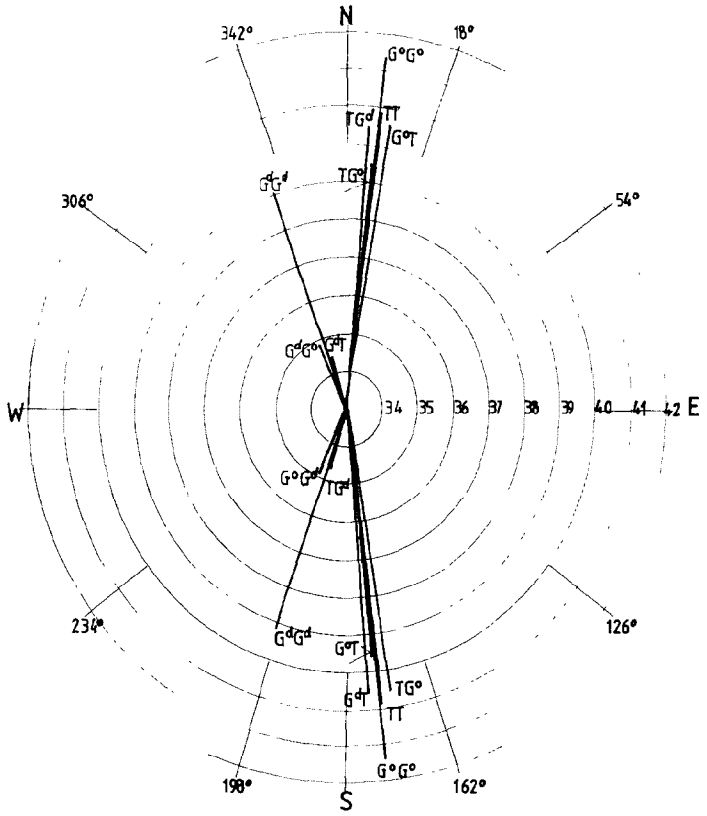

Fig. 3. The τ_m and P pseudorotational parameters of the optimised, final conformational states of the ring of **1**. There is a W–E mirror symmetry.

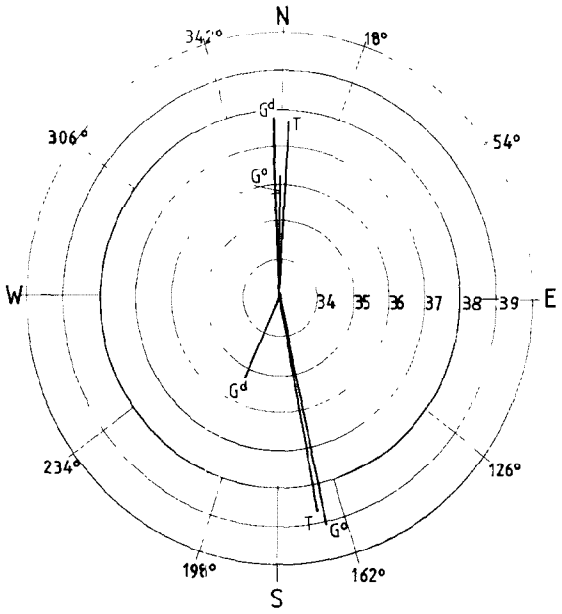

Fig. 4. The τ_m and P pseudorotational parameters of the final conformational states of the ring of **2**.

The optimum conformations defined by the pseudorotation parameters P and τ_m are described for the different hydroxyl orientations in Figs. 3 and 4. They are characterised by their total energy E_{tot}, total energy corrected for zero-level vibrations E_{tot}(0 K) (so-called zero-point energy), and free enthalpy $G°$(300 K), which are entered in Tables I and II. The translational-rotational contributions to the free enthalpy differences are below 0.17 kJ.mol^{-1}, and therefore only oscillation contributions have been included.

Analysis of energy parameters of the final conformational states. — The data in Tables I and II indicate that the energy minima are well defined. Energy differences between the conformers are of the order of barriers to pseudorotation between N and S states for deoxyribose and ribose in nucleosides[7,8]. The differences $\Delta G°$(300 K) are, in general, 0.2–2.4 kJ.mol^{-1} lower than the corresponding ΔE_{tot}, with G^dG^d and G^dT conformers the only exceptions (1.4 and 0.25 kJ.mol^{-1} higher, respectively). The zero-point energy differences ΔE_{tot} (0 K) resemble $\Delta G°$(300 K) more than ΔE_{tot}.

The thermodynamic parameters of the ring of **2** show less variation than values given for the ring of **1**. This is evidently due to interactions between the vicinal hydroxyl groups, which occur only in **1**.

Equilibrium distribution of the conformers of **1** *and* **2**. — The $\Delta G°$(300 K) values can be employed to calculate the statistical weights of all possible conformers. If i denotes a conformational state of a molecule, then the relative ratio of this conformer with respect to the most stable conformation (used as a reference) is given by a modification of Eq. 6:

TABLE I

DIFFERENCES (kJ.mol^{-1}) IN TOTAL ENERGY ΔE (TOP), ZERO POINT ENERGY E(0 K) (MIDDLE), AND FREE ENTHALPY $\Delta G°$(300 K) (BOTTOM) FOR THE EQUILIBRIUM CONFORMERS OF THE RING OF **1**[a]

3	G^d	T	$G°$
2			
		7.32	10.40
$G°$	~58.0[b]	6.68	9.14
		6.42	7.95
	10.28	12.92	10.42
T	10.69	12.61	9.48
	10.10	12.20	8.29
	5.24	4.82	0.0
G^d	6.16	5.09	0.0
	6.68	5.07	0.0

[a]Energetic values for the S states are obtained by permutation of the *2* and *3* symbols. [b]The local energy minimum has not been found. The value has been estimated without an optimisation procedure.

TABLE II

DIFFERENCES (kJ.mol^{-1}) IN TOTAL ENERGY ΔE (TOP), ZERO-POINT ENERGY E(0 K) (MIDDLE), AND FREE ENTHALPY $\Delta G°$ (300 K) (BOTTOM) FOR THE EQUILIBRIUM CONFORMERS OF THE RING OF **2**

	G^d	T	$G°$
	7.17	2.37	4.17
N-states	6.92	1.14	2.78
	6.03	0.88	2.28
	0.0	6 32	8 15
S-states	0.0	5.83	7 58
	0.0	4.72	6.86

TABLE III

MOLAR RATIOS x_N OF THE N CONFORMERS[a]

Compound	*I*	*II*	*III*
	0.50	0.50	0.94
1	0.50	0.50	0.92
	0.50	0.50	0.90
	0 36	0.84	0.83
2	0.47	0.88	0.87
	0.49	0.85	0.86

[a]The populations were calculated using the ΔE (top), ΔE(0 K) (middle), and $\Delta G°$(300 K) (bottom) values (Tables I and II) when averaging. $x_S = 1 - x_N$. I, No geometrical restrictions imposed on the hydroxyl positions; II, the HO-2 G^d orientation in the N state and the HO-3 G^d orientation in the S state have not been included in the statistical averaging procedure (these are the conformations where a weak hydrogen-bond type interaction of the hydroxyl groups with O-1 is possible); III, the HO-3 group is in the $G°$ orientation.

$$\sigma_i = \exp(-\Delta G_i^\circ/RT), \tag{7}$$

and the molar ratio of, say, the N conformers in the whole population is

$$X_N = \sum_{i\in N} \sigma_i / \left(\sum_{i\in N} \sigma_i + \sum_{j\in S} \sigma_j \right), \tag{8}$$

$$\text{and } X_S = 1 - X_N. \tag{9}$$

The results of these calculations, with either total or zero-point energies or free enthalpies used as energy term, are given in Table III. In column I of Table III, $X_N = 0.5$ for the ring of **1** because the N and S states are equally favored due to symmetry. This does not hold for the ring of **2** and the S conformer becomes more probable, with significant dependence on the functions used [0.36 kJ.mol^{-1} for ΔE_{tot}, 0.49 for ΔG°(300 K)]. Since ΔG° values determine an equilibrium constant, this means that, with no geometrical restrictions imposed on orientations of hydroxyl group(s), $X_N \cong X_S \cong 0.5$.

This conclusion should be compared with that of previous force-field studies[8] which assumed an effective potential for the interaction of the hydroxyl groups and were based on slightly different molecular models. Classical energetic analysis (equation *4*) yielded $\sigma_N = 0.48$, $\sigma_S = 0.51$, $\sigma_E = 0.01$ for ribose, and $\sigma_N = 0.11$, $\sigma_S = 0.74$, $\sigma_E = 0.11$ for 2′-deoxyribose moieties. Comparing these data with the entries in column I of Table III, it appears that ΔE_{tot} determines qualitatively the N $\rightleftharpoons$ S equilibrium. Our results indicate, however, that the oscillation corrections are responsible for the shift of the traditionally accepted equilibrium constant.

Because there are no experimental results on the N $\rightleftharpoons$ S equilibrium in the vapour state, the theoretical results cannot be compared directly with measured data. A rough estimate based on crystallographic structure analysis[3] for ribonucleosides gives $X_N \cong X_S \cong 0.5$; for deoxyribonucleosides, $X_N \cong 0.2$ and $X_S \cong 0.8$. In recent ^{1}H-n.m.r. studies of solutions of unsubstituted furanoses and their phosphates in D_2O, the preferred ring pucker was established as C-3-*endo* (N) for both ribo and deoxyribo derivatives[4].

If hydroxyl groups are oriented preferentially "away" from the sugar in order

TABLE IV

MEAN FREE-ENTHALPY DIFFERENCES $\overline{\Delta G_{NS}}$ (300 K) (kJ.mol^{-1}) BETWEEN THE N AND S CONFORMERS OF THE RINGS OF **1** AND **2**

Compound	*I*[a]	*II*[a]	*III*[a]
1	0.00	0.00	−5.54
2	0.05	−4.33	−4.58

[a]See footnote to Table III.

to form hydrogen bonds with bulk water, the data entered in column II of Table III are obtained from corresponding energy values in Table I. Because of the symmetry of the ring of **1**, the equilibrium remains unchanged. For the ring of **2**, however, C-3-*endo* (N) is now largely preferred, a finding consistent with above-mentioned ^{1}H-n.m.r. studies.

Column III of Table III describes the equilibrium when HO-3 is in the G° orientation. This particular conformation is the same as that of an O-3–P group, if a polynucleotide is in a double-helical arrangement. The equilibrium of the ring in **1** is now largely shifted towards C-3-*endo* (N) and the behaviour of the ring of **2** is similar, but not so pronounced. These findings parallel structural results obtained with doubly helical nucleic acids which, for RNA, occur only in C-3′-*endo* form and for DNA can adopt C-3′-*endo* as well as C-2′-*endo* states, depending on humidity and salt conditions. However, it should be stressed that, on the polymer level, non-bonded interactions of substituents at C-2′ are probably more important for the stabilization of the secondary structures[20,21].

All the possible rotamers can be classified into N or S types, and the population analysis allows the mean free-enthalpy difference between the puckering modes to be estimated:

$$\overline{\Delta G_{NS}} = \overline{G_N} - \overline{G_S} = RT \ln (X_N/X_S). \qquad (10)$$

Results obtained from such calculations are collected in Table IV. The N and S states are equally probable for the rings of the model compounds **1** and **2** . If limitations are imposed on the orientation of the hydroxyl groups to mimic the behaviour in aqueous solution or in a double helix, the C-2-*endo* (S) pucker is destabilised by up to 5.5 kJ.mol^{-1}.

Because the optimisation procedure employing the gradient method does not significantly alter the relative orientations of the hydroxyl groups, it may be concluded that a unique pseudorotational path (reaction-path coordinate) does not exist for the N ⇆ S transition. There are, in fact, as many paths as there are favourable configurations of the hydroxyl groups. In order to find the energy and geometrical profiles along a path, all of the degrees of freedom of a furanose moiety must be optimised for varying P values.

Only qualitative information is available presently, assuming that such a path can be approximated by computer modelling based on geometrical considerations[18]. Thus, for the ring of **1**, two paths of opposite behaviour can be envisaged, one characterised by the TT configuration of the hydroxyl groups and the second by G^{d}G^o, with the same geometry of the ring system adopted in both cases.

The energy profiles corresponding to these two cases are presented in Fig. 5. In Fig. 5A, the profile is rather "regular" and reminiscent of those published[2–8]. The hydrogen atoms are oriented opposite to each other and, at P = 90° and P = 270°, the molecule is mirror-symmetrical. The second profile is unusual because there is steric hindrance between the hydroxyl groups in the S-state. It seems that

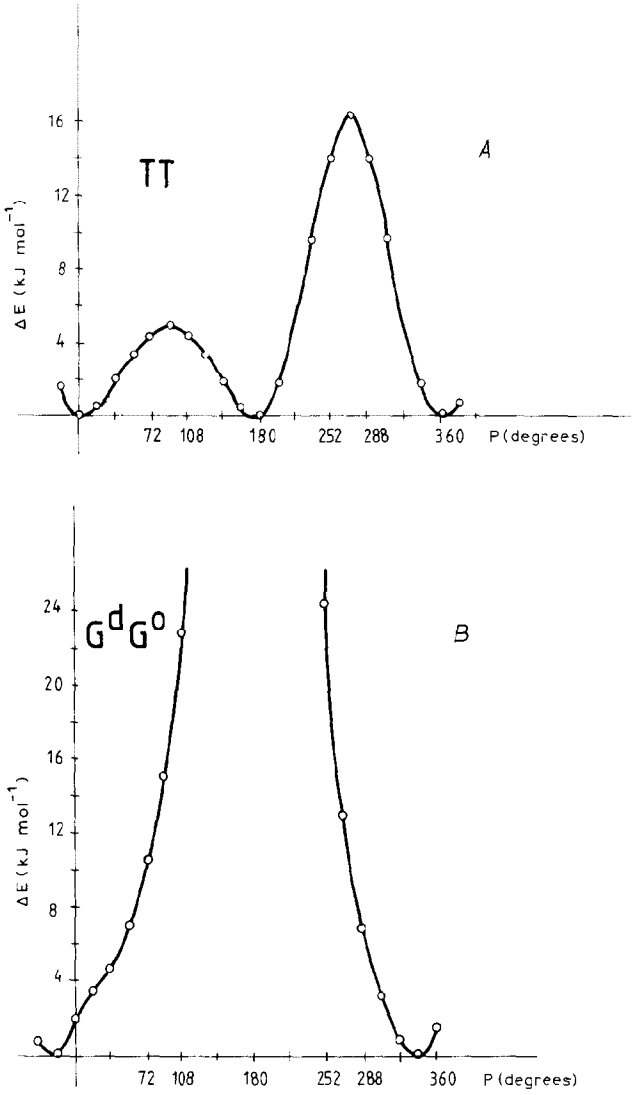

Fig. 5. Energy profiles along an approximate pseudorotational path, with the configurations of the hydroxyl groups being *A*, TT; and *B*, G^dG^o.

the conformations located on this path are highly improbable, although the energy minimum of the N state is the lowest one (G^dG^o is 12.9 kJ.mol^{-1} below TT in Table I). The stabilisation of N is due to antiparallel orientation of the hydroxyl groups and simultaneous weak attraction between HO-2 and O-1. This example suggests that the N $\rightleftarrows$ S interconversion of furanose rings is probably more complicated than previously assumed.

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