Microwave Spectroscopy

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Six Pyranoside Forms of Free 2-Deoxy-D-ribose**

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Carbohydrates are one of the most versatile biochemical building blocks, widely acting in energetic, structural, or recognition processes.[1] The interpretation of the biological activity of saccharides is based on the structure and relative stability of their conformers. One of the obstacles to resolving the basic structure issues arises from their ability to form strong intermolecular hydrogen bonds with polar solvents, which in turn can result in conformational changes. A clear picture of the conformational panorama of isolated 2-deoxy-D-ribose has been revealed using Fourier-transform microwave spectroscopy in conjunction with a UV ultrafast laser ablation source. Additionally, the availability of rotational data has been the main bottle-neck for examining the presence of these building blocks in interstellar space,[2] so these studies could also be useful to the astrochemistry community.

2-Deoxy-D-ribose (2DR, $C_5H_{10}O_4$; Figure 1a) is an important naturally occurring monosaccharide, present in nucleotides, which are the building blocks for DNA.[3] In DNA, 2DR is present in the furanose (five-membered) ring form, whereas free in aqueous solution it cyclizes into five- or six-membered rings, with the latter—the pyranoid form—being dominant.[4] By closing the chain into a six-membered ring, the C_1 carbon atom is converted into an asymmetric center, yielding two possible stereochemical α and β anomeric species (Figure 1b). In aqueous solution, 2DR primarily exists as a mixture of nearly equal amounts of α and β pyranose forms, present in their low-energy chair conformations, $^4C_1$ and $^1C_4$ (Figure 1c).[4] Such configurations are connected through ring inversion, thus establishing the axial or equatorial position of the OH group for each conformer. In addition, the monossacharides exhibit an unusual preferential stabilization of pyranose rings containing an axial OH group at the C_1 carbon over the equatorial orientation, widely known as the anomeric effect,[5] although its physical origin remains controversial.[6] Nevertheless, structural analysis of 2DR must take into consideration the intramolecular hydrogen bonding between adjacent OH groups. The formation of hydrogen-bond networks reinforces their stability owing to hydrogen-bond cooperativity effects.[7] Such networks are fundamental to the molecular recognition of carbohydrates.[8] By dissecting all these factors we can determine the most stable conformers of 2DR and the relative arrangement of the different hydroxy groups under isolated conditions, such as in the gas phase.

In vacuo theoretical calculations, carried out on α-/β-pyranoses, α-/β-furanoses, and open-chain conformations, predict 15 furanose and pyranose forms (Figure 1d, Table 1) in an energy window of 12 kJ mol^{-1} above the predicted cc-α-pyr $^4C_1$ global minimum. The notation used to label the different conformers include the symbols α and β to denote the anomer type, $^4C_1$ and $^1C_4$ to denote the pyranose chair form, C2-endo or C3-endo to denote the furanose envelope forms, and "c" or "cc" to indicate a clockwise or counterclockwise configuration of the adjacent OH bonds, respectively. A number is added to provide the MP2 energy ordering within the same family. To validate the predicted conformational behavior, comparison with precise experimental data of 2DR is needed. Previous experiments to determine the conformation of monosaccharides were based on X-ray and NMR measurements.[9,4] However, these data are influenced by environmental effects associated with the solvent or crystal lattice. Recently, an IR spectrum of 2DR in an inert matrix in

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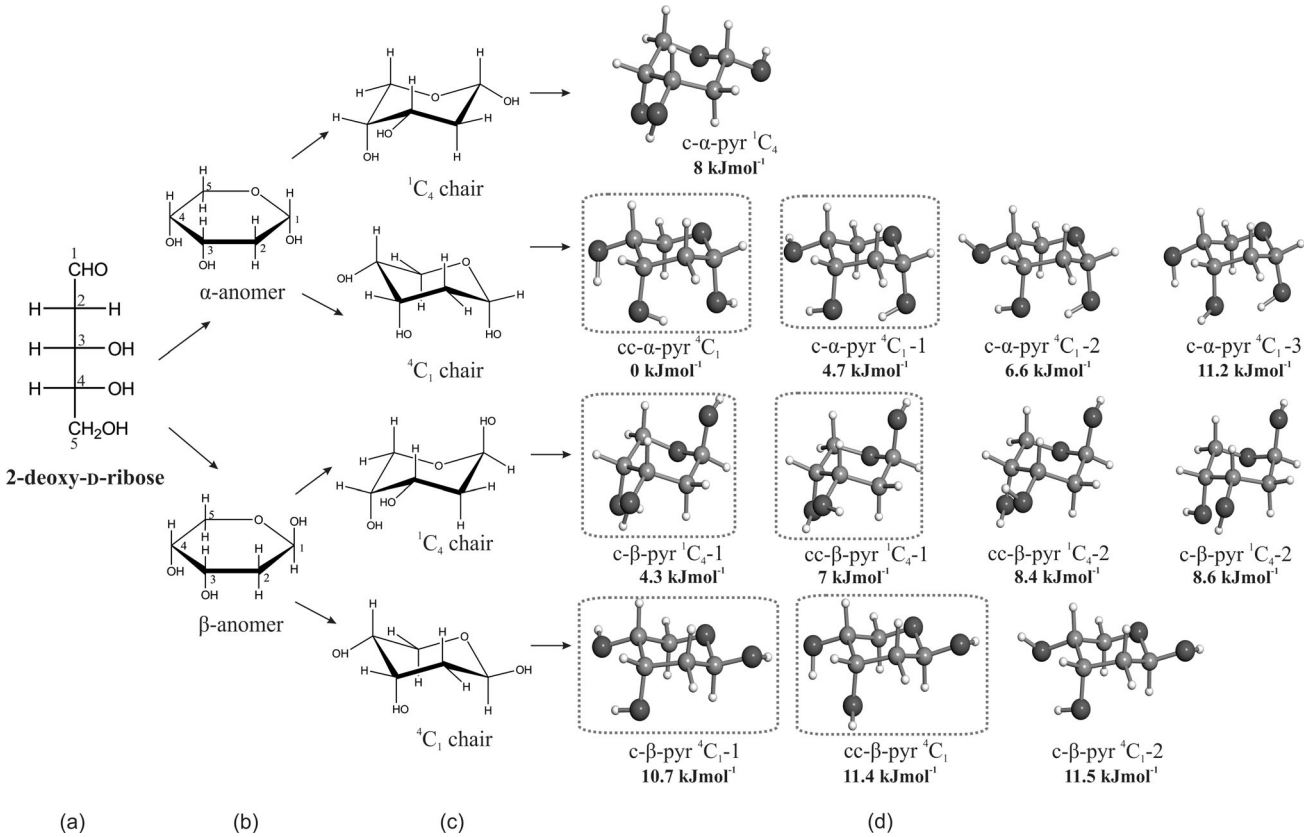

Figure 1. a) Fisher projection of 2-deoxy-D-ribose. b) Haworth projections of α and β anomers. c) $^1C_4$ and $^4C_1$ chair conformations. d) Predicted conformers within 12 kJ mol^{-1} from MP2(full)/6-311++G(d,p) ab initio computations; the observed conformers are in dashed boxes. H white; C light gray; O dark gray.

the region of the stretching OH vibration was interpreted by summing the modeled spectra for several α and β conformers.[10] Gas-phase studies of some phenyl-substituted monosaccharides,[11a] polysaccharides,[11b] or sugar complexes[11c] have been investigated by UV and IR double-resonance hole-burning spectroscopy experiments, but no data have been reported for the corresponding 2DR derivative.

Table 1: Spectroscopic parameters and relative energies for the α and β lowest-energy conformers of 2-deoxy-D-ribose.[a]

	A[b] [MHz]	B [MHz]	C [MHz]	$\lvert\mu_a\rvert$ [D]	$\lvert\mu_b\rvert$ [D]	$\lvert\mu_c\rvert$ [D]	ΔE[c] [kJ mol^{-1}]	ΔG[d] [kJ mol^{-1}]
cc-α-pyr $^4C_1$	2492	1533	1250	2.9	0.3	0.3	0	0
c-α-pyr $^4C_1$-1	2518	1535	1259	3.0	0.6	1.4	4.7	4.7
c-α-pyr $^4C_1$-2	2512	1531	1258	3.6	0.7	2.8	6.6	6.0
c-α-pyr $^1C_4$	2503	1395	1077	1.8	1.6	1.9	8.0	6.7
c-α-pyr $^4C_1$-3	2511	1520	1249	1.0	3.3	1.6	11.2	10.6
c-β-pyr $^1C_4$-1	2447	1527	1158	2.6	1.2	0.5	4.3	3.3
cc-β-pyr $^1C_4$-1	2463	1524	1150	1.0	2.1	0.6	7.0	5.6
cc-β-pyr $^1C_4$-2	2446	1528	1150	0.2	1.1	2.2	8.4	6.8
c-β-pyr $^1C_4$-2	2455	1522	1154	1.2	2.6	0.3	8.6	6.7
c-β-pyr $^4C_1$-1	2956	1279	1030	1.7	2.0	0.2	10.7	8.9
cc-β-pyr $^4C_1$	2948	1273	1028	0.2	2.2	1.0	11.4	9.4
c-β-pyr $^4C_1$-2	2947	1279	1029	2.2	0.6	1.1	11.5	9.4
cc-α-fur-C_2-*endo*-1	2527	1379	1155	0.6	2.6	0.6	6.7	3.5
cc-α-fur-C_2-*endo*-2	2627	1261	1035	1.3	2.0	1.5	10.2	6.5
cc-α-fur-C_2-*endo*-3	2576	1371	1172	1.9	2.0	0.5	12.0	8.5

[a] Within 12 kJ mol^{-1} at MP2(full)/6-311++G(d,p). [b] A, B, and C represent the rotational constants, $\lvert\mu_a\rvert$, $\lvert\mu_b\rvert$, and $\lvert\mu_c\rvert$ are the absolute values of electric dipole moment components (1 D = 3.3356×10^{-30} C m). [c] Electronic energies including zero-point energy correction. [d] Gibbs energies calculated at 298 K.

The intrinsic difficulty of working with gas-phase 2DR (or carbohydrates, in general) is due to the labile nature of the solid sample (m.p. = 89–90 °C) and inherent difficulties for vaporization. A number of powerful strategies have been found, which use a combination of laser ablation for transferring intact molecules into the gas phase, rapid cooling in a free jet expansion to stabilize their conformers, and highly selective Fourier-transform microwave spectroscopy to probe the most stable conformers (this method is known as LA-MB-FTMW).[12] Structural studies of amino acids,[13] nucleic acid bases,[14] neurotransmitters,[15] drugs (like aspirin),[16] and sugars such as glucose[17] benefit from this LA-MB-FTMW technique.[12] Recently, the microwave spectra of ribose[18] and fructose[19a] have been characterized using a UV ultrafast laser ablation source with a Balle–Flygare FTMW spectrometer. In the last years, new broadband Fourier-transform microwave techniques[20] have allowed for rapid acquisition of the rotational spectrum in wide frequency ranges. Recently, a picosecond laser ablation source has been assembled for these techniques[21,22] at the University of Valladolid and applied to the conformational studies of vitamin C[22] and D-fructose.[19b] The spectrum of 2DR was observed and assigned independently in Valladolid and Bilbao using a chirped-pulse (CP) FTMW technique and a Balle-Flygare FTMW spectrometer, respectively, in both cases combined with a UV picosecond laser source (for more details see the Experimental Section of the Supporting Information). Six different rotameric species (labeled I to VI) were identified, after the lines known to belong to photo-fragmentation species[19b] were removed from the broadband spectra recorded in Valladolid (see Figure 2). Assignments were mainly based on the identification of characteristic patterns of μ_a-R-branch progressions in the 6–12 GHz frequency range. The rotational constants collected in Table 2 (see the complete results in Table S1) were determined by a Watson semirigid rotor Hamiltonian[23] of the measured transitions (Supporting Information, Tables S2–S7).

A comparison of the experimentally determined values (Table 2) with those predicted ab initio (Table 1) enabled the identification of the six detected rotamers as particular conformers of 2DR. All structures observed were α/β-pyranoses forms. We found no evidence of either α/β-furanoses or any linear forms in the gaseous 2DR. The conformational assignment used the rotational constants, type, and magnitude of the observed spectrum and the relative intensity of the microwave transitions. The experimental rotational constants of rotamers I and II are only consistent with those predicted for the conformers c-β-pyr $^1C_4$-1 and cc-β-pyr $^1C_4$-1. However, their absolute values do not allow discrimination between them. Considering that one departs from the clockwise orientation of the OH groups of the c-β-pyr $^1C_4$-1 conformer towards the cc-β-pyr $^1C_4$-1 conformer counterclockwise orientation, the predicted changes in rotational constants ($\Delta A = -16$ MHz, $\Delta B = 3$ MHz, and $\Delta C = 8$ MHz) are in good agreement with those calculated from experimental values ($\Delta A = -11.7$ MHz, $\Delta B = 2.4$ MHz, and $\Delta C = 7.5$ MHz), thus allowing us to assign rotamer I as conformer c-β-pyr $^1C_4$-1 and rotamer II as conformer cc-β-pyr $^1C_4$-1. Also, the intensities of the measured transitions are in agreement with the predicted values of the dipole moment components and the selection rules, further supporting this assignment. Analogously, the rotational constants of rotamers III and IV should be related to either c-β-pyr $^4C_1$-1 or cc-β-pyr $^4C_1$ conformers. Only μ_a- and μ_b-type spectra for rotamer III and μ_b- and μ_c-type spectra for rotamer IV were observed. Based on the predicted values of the dipole moment components, rotamer III can only be ascribed as c-β-pyr $^4C_1$-1, whereas rotamer IV is cc-β-pyr $^4C_1$. For rotamers V and VI, rotational constants are consistent with those predicted for α $^4C_1$ pyranoses. Again, the selection rules and intensities observed indicate that rotamers V and VI should be assigned to conformers cc-α-pyr $^4C_1$ and c-α-pyr $^4C_1$, respectively. The fact that we did not observe conformers c-α-pyr $^4C_1$-2, cc-β-pyr $^1C_4$-2, c-β-pyr $^1C_4$-2, and c-β-pyr $^4C_1$-2 in Table 1 can be safely attributed to a collisional relaxation in the jet,[24] because it differs from the observed conformers only in the orientation of one of the hydroxy groups at C_3 or C_4 (see some examples of the calculated interconversion barriers in the Supporting Information, Figures S1,S2).

The population ratios for α and β conformers—c-β-pyr $^1C_4$-1(I):cc-β-pyr $^1C_4$-1(II):c-β-pyr $^4C_1$-1(III):cc-β-pyr $^4C_1$ (IV):cc-α-pyr $^4C_1$(V):c-α-pyr $^4C_1$-1(VI) = 1:0.06(1):0.38(4):0.11(1):-0.15(1):0.02(2)—have been estimated from the transition intensities,[25] taking into account the values of dipole moment components from Table 1 and by assuming that the cooling in the supersonic expansion brings all the molecular systems to the lowest vibrational state of each observed conformer. Prior to the interpretation of these data, although the

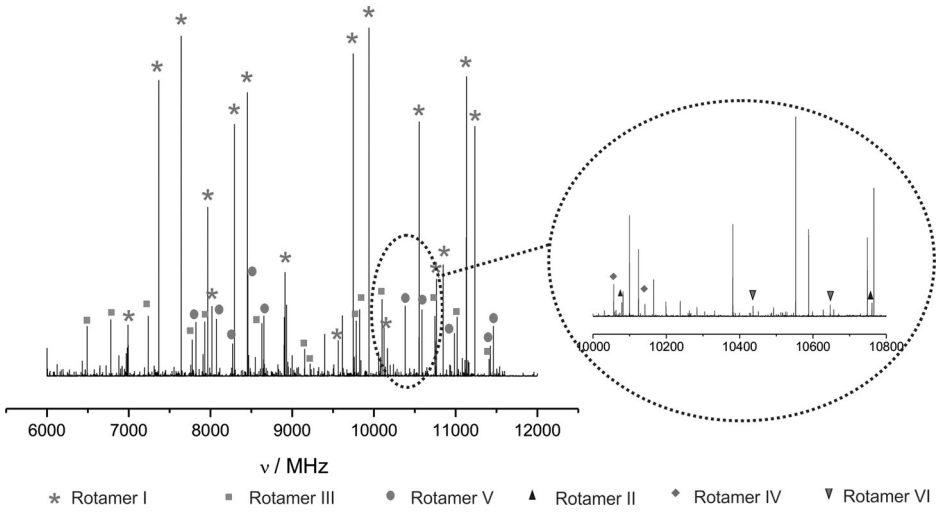

Figure 2. Broadband microwave spectrum of 2-deoxy-D-ribose.

Table 2: Experimental spectroscopic parameters for the six observed conformers of 2-deoxy-D-ribose.

Parameter	Rotamer I c-β-pyr $^1C_4$-1	Rotamer II cc-β-pyr $^1C_4$-1	Rotamer III c-β-pyr $^4C_1$-1	Rotamer IV cc-β-pyr $^4C_1$	Rotamer V cc-α-pyr $^4C_1$	Rotamer VI c-α-pyr $^4C_1$-1
A[a] [MHz]	2437.82389(32)[e]	2449.4937 (10)	2934.15899 (60)	2921.37984 (69)	2484.4138 (40)	2505.0150 (12)
B [MHz]	1510.72826 (24)	1508.31836 (62)	1271.16943 (40)	1266.99318 (79)	1517.76532 (26)	1521.47507 (52)
C [MHz]	1144.98038 (27)	1137.47992 (43)	1022.34862 (35)	1020.28439 (76)	1238.99757 (27)	1246.45004 (53)
μ_a[b]	Obs[f]	Obs	Obs	–	Obs	Obs
μ_b	Obs	Obs	Obs	Obs	–	–
μ_c	Obs	Obs	–	Obs	–	Obs
σ[c] [KHz]	5.2	7.3	5.0	7.0	1.8	4.0
N[d]	67	21	43	20	19	16

[a] A, B, and C are the rotational constants. [b] Electric dipole moment. [c] RMS deviation of the fit. [d] Number of fitted transitions. [e] Standard error in parenthesis in the units of the last digit. [f] Obs = observation of a-, b-, and c-type transitions for each structure.

cc-α-pyr $^4C_1$ conformer has been predicted to be 4.3 kJ mol^{-1} more stable than the c-β-pyr $^1C_4$ one, it is only reasonable to assume that the 2DR rotational spectra will reflect the composition of the α and β forms in the solid commercial sample. The interconversion between the α and β anomers is a solvent-mediated reaction and would not occur that easily during evaporation,[26] especially if the sample is completely dry.[27] Hence, our results indicate that 2DR exists in the gas phase as a mixture of approximately 10 % of α and 90 % of β pyranose forms, thus showing the dominance of the β-$^1C_4$ pyranose form, as found in the previous X-ray crystal study.[9] The relative hypothetical equilibrium populations for the β forms—c-β-pyr $^1C_4$-1(I):cc-β-pyr $^1C_4$-1(II):c-β-pyr $^4C_1$-1-(III):cc-β-pyr $^4C_1$(IV) = 1:0.39:0.11:0.08, predicted from the Gibbs energies—are not in total agreement with the experimental abundances we found, particularly in the case of the pair cc-β-pyr $^1C_4$(II):c-β-pyr $^4C_1$(III). This must be due to either to an incorrect evaluation of the ab initio energies, collisional relaxation of high-energy conformers to low-energy conformers,[24] or the result of a series of processes that include the laser-vaporization of the solid.[28] In this way, the relative observed population ratio can be only tentatively related to a population distribution close to those of equilibrium at the temperature of the carrier gas, assuming that a high collisional rate exists in the seeding region.[29] Nevertheless, our experimental results reflect the most abundantly observed α and β anomers, which are predicted as the global minimum.

In Bilbao, with the Balle-Flygare FT-MW technique, thanks to the high sensitivity, we were able to extend the spectral measurements to all five monosubstituted ^{13}C species and the endocyclic ^{18}O species using their natural abundance (approximately 1.1 % and 0.2 %) for the most abundant c-β-pyr $^1C_4$-1 conformer (see Tables S8–S10). The isotopic information was used to derive substitution and effective structures for this species, shown in Table S11 and an interactive 3D model in the Supporting Information.

The detected conformers of 2DR, depicted in Figure 3, can be rationalized in terms of factors that may contribute to their stabilization. The two observed α conformers, cc-α-pyr $^4C_1$ and c-α-pyr $^4C_1$-1, are stabilized by the anomeric effect; they have a $^4C_1$ ring configuration, thus leading the anomeric OH group towards the axial position. The hydroxy groups of both conformers are located at the same side of the ring and are able to form chains of hydrogen bonds, which in turn, are strongly reinforced by sigma hydrogen-bond cooperativity.[7] The most abundant α form cc-α-pyr $^4C_1$ presents a counter-clockwise arrangement of the OH groups with a chain of three hydrogen bonds $O_{(4)}H\cdots O_{(3)}H\cdots O_{(1)}H\cdots O_{ring}$, while the less abundant c-α-pyr $^4C_1$-1 shows a chain of two hydrogen bonds $O_{(1)}H\cdots O_{(3)}H\cdots O_{(4)}H$. The anomeric effect in the most abundant β form c-β-pyr $^1C_4$-1 is reinforced by an intramolecular hydrogen bond network $O_{(3)}H\cdots O_{(4)}H\cdots O_{ring}$. Conformers c-β-pyr $^4C_1$-1 and cc-β-pyr $^4C_1$, with the anomeric hydroxy group in the equatorial position, are stabilized by two non-cooperative intramolecular hydrogen bonds. All the conformers exhibit a mutual gauche configuration for the hydroxy groups at the C_3 and C_4 positions, thus establishing that the gauche effect is not a discriminating factor for the stability. Other kinds of stabilization, such as the Hassel–Ottar and delta-two effects, play a secondary role in monosaccharides.[30]

Compared to ribose the absence of the hydroxy group at C_2 in 2-deoxyribose limits the possibility of forming hydrogen bonds and in practice leads to a weakening of the cooperative hydrogen-bond network, altering the relative abundances. For example, the most stable α-pyranose form c-α-pyr $^1C_4$ of ribose has not been detected in 2DR. The absence of an $O_{(2)}H$ group reverses the arrangement of the OH groups in the most

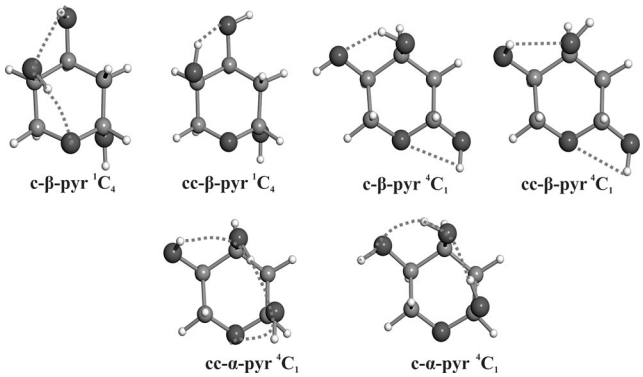

Figure 3. The six observed conformers of 2-deoxy-D-ribose showing the intramolecular hydrogen bond arrangements (dashed lines). H white; C light gray; O dark gray.

stable β-pyranose forms (from clockwise c-β-pyr $^1C_4$ of 2DR to counter-clockwise in ribose cc-β-pyr $^1C_4$) to maximize the number of hydrogen bonds (two in cc orientation versus one in the clockwise arrangement).

In summary, the anomeric effect and hydrogen bonding are the main factors controlling the conformational behavior of isolated 2DR, represented by two α- and four β-pyranose conformers. Hydroxy groups are preferentially orientated in such way to yield cooperative hydrogen bonding as efficiently as possible. When 2DR is vaporized, it exists predominantly in the pyranose form with a relative abundance of 10% of α and 90% of β forms in accordance with its crystalline composition. In this context, the experimental ionization energy of 9.1 eV of gas-phase 2DR, obtained using tunable vacuum UV synchrotron radiation,[31] which has been previously ascribed to α pyranose forms, should actually correspond to the β forms.

The question remains as to how solvation affects the equilibrium between the pyranose and furanose forms. The evidence collected so far supports that pyranoses are more stable both in gas-phase and solution, so the presence of ribose and deoxyribose in RNA or DNA cannot be merely attributed to a preference for furanoses in a physiological medium. Some structural arguments could be based on the existence of an exocyclic hydroxymethyl group (at C-5) in the furanose form. Both DNA and RNA involve a phosphate-linked chain, connected through bonds between the (exocylic) CH_2OH OH-5′ and (cyclic) OH-3′ groups. This could not occur if the ribose/deoxyribose units were in the pyranose form. With this argument the evolutionary preference might be structural and connected to the availability of the "CH_2O-" linker. The question of why DNA/RNA chains grow by way of OH3′ and OH5′ could then be an option of suitability, because there are several combinations of hydroxy groups that can build biologically useful shapes.

Alternatively, Eschenmoser[32,33] has suggested that the use of RNA in biology is not due to base-paring strength, but to high tolerance of base-pair mismatches. However, the question remains open because no experiment has yet shown a nucleotide with a reasonable ability to replicate non-enzymatically under "natural conditions".[34]

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[1] a) P. Colins, R. Ferrier, *Monosacccharides: Their Chemistry and Their Roles in Natural Products*, Wiley, New York, **1995**; b) W. Pigman, D. Horton, *The Carbohydrates: Chemistry and Biochemistry*, Academic Press, New York, **1972**.

[2] E. Herbst, E. F. van Dishoeck, *Annu. Rev. Astron. Astrophys.* **2009**, *47*, 427.

[3] a) J. D. Watson, F. H. C. Crick, *Nature* **1953**, *171*, 737 – 738; b) W. Saenger, *Principles of Nucleic Acid Structure*, Springer, New York, **1984**, pp. 1 – 556.

[4] a) S. J. Angyal, *Angew. Chem.* **1969**, *81*, 172; *Angew. Chem. Int. Ed. Engl.* **1969**, *8*, 157; b) M. Rudrum, D. F. Shaw, *J. Chem. Soc.* **1965**, 52; c) R. U. Lemieux, J. D. Stevens, *Can. J. Chem.* **1966**, *44*, 249; d) S. J. Cortes, T. L. Mega, R. L. Van Etten, *J. Org. Chem.* **1991**, *56*, 943.

[5] a) E. Juaristi, G. Cuevas, *Tetrahedron* **1992**, *48*, 5019; b) C. L. Perrin, K. B. Armstrong, M. A. Fabian, *J. Am. Chem. Soc.* **1994**, *116*, 715.

[6] a) E. J. Cocinero, P. Çarçabal, T. D. Vaden, J. P. Simons, B. G. Davis, *Nature* **2011**, *469*, 76; b) M. P. Freitas, *Org. Biomol. Chem.* **2013**, *11*, 2885; c) C. Wang, Z. Chen, W. Wu, Y. Mo, *Chem. Eur. J.* **2013**, *19*, 1436; d) G. F. Bauerfeldt, T. M. Cardozo, M. S. Pereira, C. O. da Silva, *Org. Biomol. Chem.* **2013**, *11*, 299.

[7] a) M. López de la Paz, G. Ellis, M. Pérez, J. Perkins, J. Jiménez-Barbero, C. Vicent, *Eur. J. Org. Chem.* **2002**, 840; b) G. A. Jeffrey, W. Saenger, *Hydrogen Bonding in Biological Structures*, Springer, New York, **1991**.

[8] a) G. A. Jeffrey, *Food Chem.* **1996**, *56*, 241; b) C. A. Hunter, *Angew. Chem.* **2004**, *116*, 5424; *Angew. Chem. Int. Ed.* **2004**, *43*, 5310; c) N. S. Greenspan, *Curr. Top. Microbiol. Immunol.* **2001**, *260*, 65.

[9] S. Furberg, *Acta Chem. Scand.* **1960**, *14*, 1357.

[10] T. Yu. Nikolaenko, L. A. Bulavin, D. N. Govorun, *J. Appl. Spectrosc.* **2011**, *78*, 751.

[11] a) P. Çarçabal, R. A. Jockusch, I. Hünig, L. C. Snoek, R. T. Kroemer, B. G. Davis, D. P. Gamblin, I. Compagnon, J. Oomens, J. P. Simons, *J. Am. Chem. Soc.* **2005**, *127*, 11414, and references therein; b) E. J. Cocinero, D. P. Gamblin, B. G. Davis, J. P. Simons, *J. Am. Chem. Soc.* **2009**, *131*, 11117; c) E. J. Cocinero, P. Çarçabal, T. D. Vaden, B. G. Davis, J. P. Simons, *J. Am. Chem. Soc.* **2011**, *133*, 4548.

[12] a) J. L. Alonso, C. Pérez, M. E. Sanz, J. C. López, S. Blanco, *Phys. Chem. Chem. Phys.* **2009**, *11*, 617, and references therein; b) E. J. Cocinero, A. Lesarri, P. Écija, J.-U. Grabow, J. A. Fernández, F. Castaño, *Phys. Chem. Chem. Phys.* **2010**, *12*, 12486.

[13] I. Peña, M. E. Sanz, J. C. López, J. L. Alonso, *J. Am. Chem. Soc.* **2012**, *134*, 2305, and references therein.

[14] J. L. Alonso, V. Vaquero, I. Peña, J. C. López, S. Mata, W. Caminati, *Angew. Chem.* **2013**, *125*, 2387; *Angew. Chem. Int. Ed.* **2013**, *52*, 2331, and references therein.

[15] C. Cabezas, I. Peña, J. C. López, J. L. Alonso, *J. Phys. Chem. Lett.* **2013**, *4*, 486, and references therein.

[16] C. Cabezas, J. L. Alonso, J. C. López, S. Mata, *Angew. Chem.* **2012**, *124*, 1404; *Angew. Chem. Int. Ed.* **2012**, *51*, 1375.

[17] M. Lozoya, C. Cabezas, S. Mata, J. C. López, J. L. Alonso in *LA-MB-FTMW Studies of Sugars*, Communication MH13, International Symposium on Molecular Spectroscopy, 66th Meeting—Ohio State University, USA, June, **2011**.

[18] E. J. Cocinero, A. Lesarri, P. Écija, F. J. Basterretxea, J.-U. Grabow, J. A. Fernández, F. Castaño, *Angew. Chem.* **2012**, *124*, 3173; *Angew. Chem. Int. Ed.* **2012**, *51*, 3119.

[19] a) E. J. Cocinero, A. Lesarri, P. Écija, A. Cimas, B. J. Davis, F. J. Basterretxea, J. A. Fernández, F. Castaño, *J. Am. Chem. Soc.* **2013**, *135*, 2845; b) C. Bermúdez, I. Peña, C. Cabezas, A. M. Daly, J. L. Alonso, *ChemPhysChem* **2013**, *14*, 893.

[20] a) G. G. Brown, B. C. Dian, K. O. Douglass, S. M. Geyer, S. T. Shipman, B. H. Pate, *Rev. Sci. Instrum.* **2008**, *79*, 053103; b) B. C. Dian, G. G. Brown, K. O. Douglass, B. H. Pate, *Science* **2008**, *320*, 924; c) J.-U. Grabow, S. Mata, J. L. Alonso, I. Peña, S. Blanco, J. C. López, C. Cabezas, *Phys. Chem. Chem. Phys.* **2011**, *13*, 21063.

[21] S. Mata, I. Peña, C. Cabezas, J. C. López, J. L. Alonso, *J. Mol. Spectrosc.* **2012**, *280*, 91.

[22] I. Peña, A. M. Daly, C. Cabezas, S. Mata, C. Bermúdez, A. Niño, J. C. López, J.-U. Grabow, J. L. Alonso, *J. Phys. Chem. Lett.* **2013**, *4*, 65.

[23] J. K. G. Watson in *Vibrational Spectra and Structure*, *Vol. 6* (Ed.: J. R. Durig), Elsevier, New York, **1977**, pp. 1 – 78.

[24] R. S. Ruoff, T. D. Klots, T. Emilsson, H. S. Gutowsky, *J. Chem. Phys.* **1990**, *93*, 3142.
[25] G. T. Fraser, R. D. Suenram, C. L. Lugez, *J. Phys. Chem. A* **2000**, *104*, 1141.
[26] P. Finch, *Carbohydrates: Structures, Syntheses and Dynamics*, Kluwer Academic Publishers, Netherlands, **1999**.
[27] L. P. Guler, Y.-Q. Yu, H. I. Kenttämaa, *J. Phys. Chem. A* **2002**, *106*, 6754 – 6764.
[28] a) R. L. Levis, *Annu. Rev. Phys. Chem.* **1994**, *45*, 483, and references therein; b) L. V. Zhigilei, P. B. S. Kodali, B. J. Garrison, *J. Phys. Chem. B* **1998**, *102*, 2845; c) L. V. Zhigilei, E. Leveugle, B. J. Garrison, Y. G. Yingling, M. I. Zeifman, *Chem. Rev.* **2003**, *103*, 321.
[29] S. Blanco, A. Lesarri, J. C. López, J. L. Alonso, *J. Am. Chem. Soc.* **2004**, *126*, 11675.
[30] a) B. Ma, H. F. Schaefer III, N. L. Allinger, *J. Am. Chem. Soc.* **1998**, *120*, 3411; b) S. Wolfe, *Acc. Chem. Res.* **1972**, *5*, 102.
[31] D. Ghosh, A. Golan, L. K. Takahashi, A. Krylov, M. A. Ahmed, *J. Phys. Chem. Lett.* **2012**, *3*, 97.
[32] A. Eschenmoser, *Science* **1999**, *284*, 2118.
[33] M. Beier, F. Reck, T. Wagner, R. Krishnamurthy, A. Eschenmoser, *Science* **1999**, *283*, 699.
[34] D. H. Lee, J. R. Granja, J. A. Martínez, K. Severin, R. Ghadiri, *Nature* **1996**, *382*, 525.

