Interactions of Sugars with the Sodium Cation. Sensitivity of the Sodium-23 Nuclear Magnetic Resonance Line Width in Pyridine Solution

Sir:

Sugars complex sodium;[1] biological implications are diverse and widespread.[2] We report concentration and temperature dependent increases in line width of the ^{23}Na NMR signal due to complexation of the sodium cation by the sugar.

The ^{23}Na line width $\nu_{1/2}$ increases greatly when small amounts of sugars, such as fructose, galactose, or glucose are codissolved with the sodium salt in pyridine. For instance, with 0.0189 M $Na^+ClO_4^-$ in pyridine at 300 K, the following equation describes the dependence upon galactose concentration:

$$\nu_{1/2}(\text{Hz}) = 3870\,(\text{galactose}) + 49 \qquad (1)$$

In order to simplify the analysis, we selected L-sorbose (Aldrich) as a sugar in which the anomeric equilibrium favors to a great extent the α form, which is virtually the only one present in water[3] or in pyridine[4] solution. Sodium perchlorate (0.1 M) is dissolved in an 80:20 mixture of pyridine and pyridine-d_5, carefully dried by refluxing over potassium hydroxide pellets and distillation under argon. Line widths are obtained using a Bruker HFX-90 spectrometer at 23.81 MHz. The broadening $\Delta\nu_{1/2}$ is a linear function of L-sorbose concentration at each temperature in the range 250–340 (Figure 1). At such low sugar concentrations, ca. 10^{-2} M, the solution viscosity remains unaffected. This broadening does not appear to originate from slow chemical exchange of Na^+ between nonequivalent sites; a graph of log $\Delta\nu_{1/2}$ as a function of reciprocal temperature is linear, i.e., indicative of fast exchange, as previously shown for the binding of Na^+ to carbohydrates in water solution by Angyal[5] using proton NMR, and by Andrasko and Forsen[6] using sodium-23 NMR relaxation times. Writing the observed line width $\nu_{1/2}$ as:[6]

$$\pi\nu_{1/2} = \frac{(Na^+)}{(Na^+)_0}\left[\pi\nu_{1/2}{}^{s} + \frac{K(\text{sorbose})}{1 + K(Na^+)}\,\pi\nu_{1/2}{}^{a}\right] \qquad (2)$$

where $(Na^+)_0$ is the molar concentration of added salt, and (Na^+) that of uncomplexed sodium ions; K is the stability constant rather than a true equilibrium constant since the activities of both partners in pyridine are unknown; $\nu_{1/2}{}^{s}$ is the line width for the sodium cation *symmetrically* solvated by n pyridine molecules, $n = 4$ being a very likely number;[7] and $\nu_{1/2}{}^{a}$ is the line width for the sodium cation *asymmetrically* solvated by $n - p$ pyridine, and q sugar molecules. These are weak complexes[5,6] and $(Na^+) \simeq (Na^+)_0$ at the sodium concentrations used in this study. Equation 2 indeed predicts the linearity of $\nu_{1/2}$ vs. (sorbose) as observed in Figure 1. A graph of $(Na^+)_0$ vs. $(\Delta\nu_{1/2})^{-1}$ is also linear ($\rho > 0.990$ for three points at each temperature) if $\Delta\nu_{1/2} = \nu_{1/2} - \nu_{1/2}{}^{s}$, with K^{-1} as the ordinate and $\nu_{1/2}{}^{a}$ (sorbose) as the slope. The results are indicated in Table I. From the small temperature dependence of K, the entropy change $\Delta S° = -38 \pm 20$ J K^{-1} mol^{-1}. Assuming one entering sugar molecule ($q = 1$), this appears to rule out any stoichiometry other than $p = 1$, i.e., one sugar has displaced one pyridine unit. Otherwise, there would be a gain in translational entropy of ca. 105–165 J K^{-1} mol^{-1} per additional pyridine displaced, by application of the Sakur-Tetrode relationship modified for the gas-to-liquid change. The temperature dependence of the line width in the complex $\nu_{1/2}{}^{a}$ (Table I) yields the apparent activation energy for reorientation of the complex in pyridine solution; the linear graph of log $\nu_{1/2}{}^{a}$ as a function of T^{-1} ($\rho = 0.989$ for five points) has a slope E_a/R, with $E_a = 4.4 \pm 1.2$ kcal mol^{-1}. In the ignorance of the quadrupolar coupling constant and of the asymmetry parameter for the sodium nucleus in this complex, not a symmetrical top, one cannot derive a correlation time for the rotational motion of this entity.

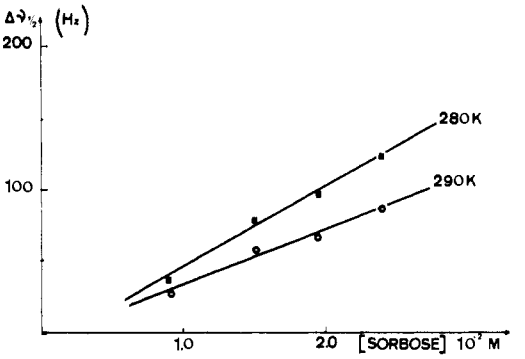


Figure 1. ^{23}Na line broadenings $\Delta\nu_{1/2}$ as a function of L-sorbose concentration at 290 K ($\rho = 0.991$) and 280 K ($\rho = 0.994$).

Table I. Interaction Parameters for the Sodium–Sugar Interactions[a]

Temp (K)	Sugar	Stability constant K (M^{-1})	Line width in complex $\nu_{1/2}{}^{a}$ (Hz)
250	Sorbose	12.5 ± 3.75	2700 ± 800
260	Sorbose	8.2 ± 2.5	1890 ± 570
270	Sorbose	7.4 ± 2.2	1640 ± 500
280	Sorbose	8.4 ± 2.5	1040 ± 310
290	Sorbose	7.2 ± 2.2	800 ± 240
300	Glucose	2.6 ± 0.8	845 ± 250
300	Galactose	5 ± 1.5	850 ± 250
300	Fructose	6.2 ± 1.8	830 ± 250

[a] The quoted uncertainties have been evaluated taking twice the standard deviation as an upper limit for the systematic error.

The stability constant for the sorbose-Na^+ complex obtained here[8] is unusually large for a sugar devoid of the characteristic *axial-equatorial-axial* arrangement of OH groups.[5] Clearly, use of pyridine as a solvent boosts the magnitude of these interactions and makes them much better amenable to study. Unfortunately, inositol, which binds strongly the Na^+ cation, is virtually insoluble in pyridine. We visualize the complex with three pyridine nitrogens and two (or three) sugar oxygens arranged around sodium. The resultant dissymmetry and electric field gradient[9] at the sodium nucleus are consistent with the extremely large $\nu_{1/2}{}^{a}$ values in Table I. Also the complex would reorient rather more slowly than the $(Na^+)_{4Pyr}$ species; the latter motion also is characterized by an activation energy of $E_a = 4.4 \pm 0.6$ kcal mol^{-1} while pyridine solvent molecules reorient with an activation energy of 2.4 kcal mol^{-1}.[11] We note furthermore that our $\nu_{1/2}{}^{a}$ values (Table I) are commensurate with those reported by Popov[12] in his pioneering study of the monensin–sodium complex.

The gratifying internal consistency of this treatment and its simplicity make it a likely explanation of the reported increases in the sodium line width. We are led to the conclusion of insertion of one sorbose molecule in the sodium solvation shell, with the attendant release of one pyridine solvent molecule.[13]

Acknowledgments. The Bruker HFX-90 FT NMR spectrometer used in this investigation was purchased with the help of a grant from Fonds de la Recherche Fondamentale Collective. We thank I.R.S.I.A. for its award of a predoctoral fellowship to C.D.

References and Notes

(1) S. J. Angyal, *Adv. Chem. Ser.*, **No. 117**, 106 (1973).
(2) C. L. Comar, F. Bronner et al., "Mineral Metabolism", Academic Press,

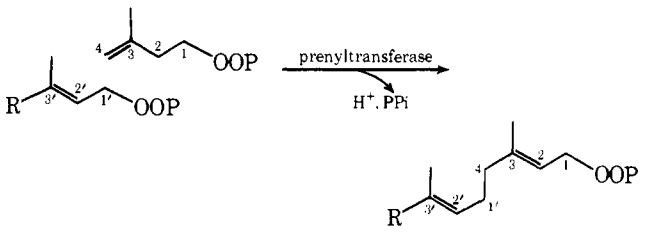
New York, N.Y., 1962; H. H. Ussing, P. Kruhoffer, J. H. Thaysen, and N. A. Thorn, The Alkali Metal Ions in Biology, *Handb. Exp. Pharmakol.*, **13** (1960).
(3) A. S. Perlin, P. H. du Penhoat, and H. S. Isbell, *Adv. Chem. Ser.*, **No. 117**, 39 (1973).
(4) More than 85% according to the ^{13}C spectrum assuming equal T_1^d values for equivalent carbons in the two anomeric forms. This anomeric distribution is not significantly changed upon addition of sodium perchlorate at the concentrations in this study.
(5) S. J. Angyal, *Tetrahedron*, **30**, 1695 (1974).
(6) J. Andrasko and S. Forsen, *Biochem. Biophys. Res. Commun.*, **52**, 233 (1973).
(7) C. Detellier and P. Laszlo, to be submitted for publication; see also A. T. Tsatsas, R. W. Stearns, and W. M. Risey, *J. Am. Chem. Soc.*, **94**, 5247 (1972).
(8) It was not possible to perform an independent check from the sodium-23 chemical shift; within experimental error, it remains invariant in the presence of sugars at the concentrations used in this study.
(9) Using a trigonal bipyramidal idealized geometry, with equatorial oxygens at 2.55 Å, and nitrogens at 2.65 Å, CNDO-2 charges of 0.25 (O) and 0.14 (N) lead to a reasonable[10] value of ca. 1 MHz for the quadrupolar coupling constant. This value could be somewhat higher if one were dealing with a sugar-bridged $Na^+ClO_4^-$ ion pair, bringing a perchlorate oxygen into van der Waals contact of the cation.
(10) D. H. Haynes, B. C. Pressman, and A. Kowalsky, *Biochemistry*, **10**, 852 (1971).
(11) J. P. Kintzinger and J. M. Lehn, *Mol. Phys.*, **22**, 273 (1971).
(12) P. G. Gertenbach and A. I. Popov, *J. Am. Chem. Soc.*, **97**, 4738 (1975).
(13) Note Added in Proof. The sodium-23 line broadenings obtained with monohydric alcohols and diols such as cyclohexane-1,1-dimethanol are negligible by comparison to those observed with sugars. Indeed the interaction is specific of sugars.

Christian Detellier, Jean Grandjean, Pierre Laszlo*
Institut de Chimie, Université de Liège
Sart-Tilman par 4000 Liège, Belgium
Received January 27, 1976

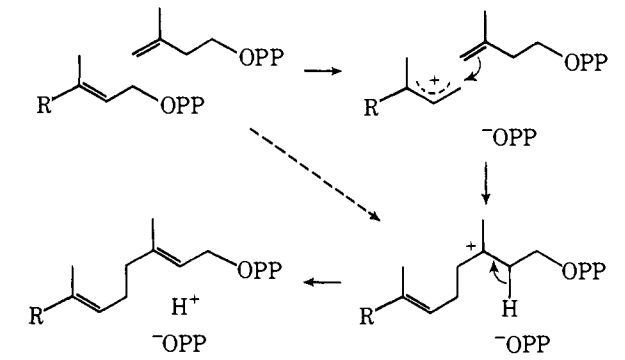
Prenyltransferase. The Mechanism of the Reaction[1]

Sir:

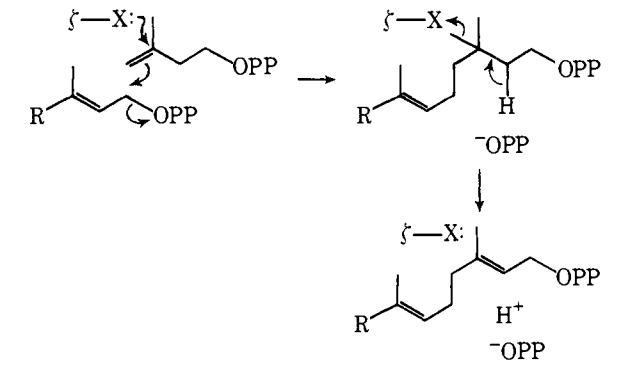
Prenyltransferase (EC 2.5.1.1) catalyzes the condensation between C_4 of isopentenyl pyrophosphate (**IPP**) and C_1' of an allylic pyrophosphate, giving the five-carbon homologue of the allylic pyrophosphate. This is the fundamental chain elongation



Scheme I. Ionization–Condensation–Elimination



Displacement–Elimination



reaction of terpene biosynthesis and leads to the formation of several important classes of natural products such as sterols, carotenoids, dolichols, and respiratory coenzymes. The mechanisms which have been proposed for prenyl transfer can be grouped into two broad categories (Scheme I): those in which condensation is initiated by heterolytic cleavage of the carbon–oxygen bond, with or without assistance from the double bond of isopentenyl pyrophosphate, yielding cationic intermediates (ionization–condensation–elimination),[2] and those in which condensation is initiated by attack of a nucleophilic group at the double bond of isopentenyl pyrophosphate with simultaneous formation of the C_1'–C_4 bond between the two substrates and rupture of the C_1'–oxygen bond (displacement–elimination).[2d,3] We reasoned that it would be possible to distinguish between the two mechanisms by replacing the R group in the allylic substrate by a trifluoromethyl group. The strong electron-withdrawing effect of a trifluoromethyl substituent ($\sigma^+ = 0.612$)[4] should retard the rate of

Table I. Effect of Methyl and Trifluoromethyl Groups

Reactant	T, °C	Solvent (% acetone–H_2O)	k	k_{CF_3/CH_3}
SN1				
E-3-Trifluoromethyl-2-buten-1-yl methanesulfonate (*E*-**1**-OMs)	20	92	5.5×10^{-8} [a-c]	1.8×10^{-6}
	20	50	1.55×10^{-6} [a,c]	
	60	50	$1.28 \pm 0.03 \times 10^{-4}$ [c]	
	70	50	$3.06 \pm 0.20 \times 10^{-4}$ [c]	
	80	50	$7.98 \pm 0.55 \times 10^{-4}$ [c]	
3-Methyl-2-buten-1-yl methanesulfonate (**4**-OMs)[d]	0	92	$3.82 \pm 0.11 \times 10^{-3}$ [c]	
	20	92	$2.99 \pm 0.12 \times 10^{-2}$ [c]	
SN2				
E-1-Chloro-4,4,4-trifluoro-2-butene (**5**)	20	100	68.8×10^{-5} [e,f]	11
E-1-Chloro-2-butene (**6**)	20	100	6.1×10^{-5} [e,g]	

[a] Extrapolated from higher temperatures. $\Delta H^{\ddagger} = 20.7$ kcal mol^{-1}, $\Delta S^{\ddagger} = -15$ eu. [b] Extrapolated 50% acetone–water, $m = 0.383$, $Y = 2.25$. [c] Units are s^{-1}. [d] $\Delta H^{\ddagger} = 15.8$ kcal mol^{-1}, $\Delta S^{\ddagger} = -12$ eu. [e] For displacement with I^-(KI), l. mol^{-1} s^{-1}. [f] E. T. McBee, R. D. Battershell, and H. P. Braendlin, *J. Am. Chem. Soc.*, **84**, 3157 (1962); J. A. Pegolotti and W. G. Young, *ibid.*, **83**, 3258 (1961). [g] L. F. Hatch and S. S. Nesbitt, *ibid.*, **73**, 358 (1951).