# A Monte Carlo Investigation of the Conformational Free Energies of the Aldohexopyranoses

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This paper is concerned with the estimation of the energy and free energy differences between the different aldohexopyranoses and between the two chair forms of each hexose. Methods have been developed to account for the deformation of the ring by steric effects between substituents. Calculated α : β ratios of the hexoses are in good agreement with experiment except for some cases in which the anomers have a large number of equatorial hydroxy groups. This effect is interpreted as evidence for an important hydration effect which results in a stabilization of anomers which fit particularly well into an ice-like lattice.

THE problem of predicting the solid state conformations and random coil (solution) dimensions of polysaccharides has attracted a good deal of attention during the last ten years. Although much progress has been made in this area,[1,2] particularly in terms of predicting general trends within and amongst different families of polysaccharides,[3] little is known about the prediction of geometries of monosaccharides or of the free energy differences between different monosaccharide conformations. This is of particular concern since random coil dimensions are sensitive to bonding geometry [4] and to small amounts of conformational impurities.[5,6] Consequently the prediction of monomer geometries and free energies is of interest both for its own sake and for its importance in determining polymer properties.

The problem of predicting the free energy differences between different monosaccharide conformations was first attacked, in a qualitative way, by Reeves [7] and later, semi-quantitatively, by Angyal.[8,9] Angyal suggested that several factors contribute to the observed free energy difference between two conformations of a monosaccharide or between two different monosaccharides, *e.g.* the number of axial groups, the number of adjacent equatorial groups, the number of 1,3-diaxial interactions, and the anomeric effect. By considering a series of experimental results he assigned free energy values to each of these terms and summed them to estimate the free energies of the 1*C* and *C*1 chair conformations of a number of monosaccharides. This approach is appealing because of its close relation to experiments, and is probably quite reliable for conformations which are not very sterically hindered. However, for a highly hindered conformation, *e.g.* the *C*1 conformation of α-D-idose, this approach will be likely to overestimate the free energy. This is because (*a*) the assumption of additivity of the effects will break down and (*b*) the magnitudes of the various effects will be decreased by deformation of the ring. Consequently, although Angyal's values are expected to be reliable for conformations of low free energy they should be treated with caution for cases involving high steric compression.

An alternative approach has been used by Rao and his co-workers.[10-13] Originally [10] they restricted their attention to calculating the minimum energies of various conformations, assuming an idealized bonding geometry and a simple semiempirical potential function, but including rotatable substituents. In later papers the potential function was extended to include coulombic terms, entropy effects were estimated, and pendant bond angles were allowed to vary for axial substituents. Although the agreement with Angyal's results is impressive some aspects of the calculations are difficult to justify. In particular the restriction of the way in which pendant bond angles were varied seems unrealistic and it would be surprising if these angles changed significantly without concomitant variation of ring angles.

More recently Rees and Smith [14] have used Metropolis-type Monte Carlo techniques [15] to estimate the average energies of the various aldopentopyranoses. They obtained excellent agreement with experimental results except for (*a*) sugars with 1,3-diaxial hydroxy groups and (*b*) sugars with almost all hydroxy groups equatorial. Since they did not allow for ring deformation it is not surprising that their calculations were less successful for sugars with strong steric compressions. The poor agreement with experiment for the type (*b*) sugars (above) is more interesting and was interpreted as evidence for a co-operative and highly selective solvent effect.[16-19] (A similar effect appears in our calculations for the aldohexopyranoses.)

[1] D. A. Rees, 'MTP Reviews of Science, Organic Chemistry, Series One,' Butterworths, London, 1973, vol. 7, p. 251.
[2] D. A. Brant, *Ann. Rev. Biophys. Bioeng.*, 1972, **1**, 369.
[3] D. A. Rees and W. E. Scott, *J. Chem. Soc.* (*B*), 1971, 469.
[4] D. A. Brant and W. F. Dimpfl, *Macromolecules*, 1970, **3**, 655.
[5] D. A. Brant and K. D. Goebel, *Macromolecules*, 1972, **5**, 536.
[6] L. G. Dunfield and S. G. Whittington, *Macromolecules*, 1974, **7**, 946.
[7] R. E. Reeves, *Adv. Carbohydrate Chem.*, 1951, **6**, 107.
[8] S. J. Angyal, *Austral. J. Chem.*, 1968, **21**, 2737.
[9] S. J. Angyal, *Angew. Chem. Internat. Edn.*, 1969, **8**, 157.
[10] V. S. R. Rao, K. S. Vijayalakshmi, and P. R. Sundararajan, *Carbohydrate Res.*, 1971, **17**, 341.
[11] K. S. Vijayalakshmi and V. S. R. Rao, *Carbohydrate Res.*, 1973, **29**, 427.
[12] K. S. Vijayalakshmi and V. S. R. Rao, *Carbohydrate Res.*, 1973, **31**, 173.
[13] K. S. Vijayalakshmi and V. S. R. Rao, *Carbohydrate Res.*, 1972, **22**, 413.
[14] D. A. Rees and P. J. C. Smith, *J.C.S. Perkin II*, 1975, 830.
[15] N. Metropolis, A. W. Rosenbluth, M. N. Rosenbluth, A. H. Teller, and E. Teller, *J. Chem. Phys.*, 1953, **21**, 1087.
[16] M. A. Kabayama and D. Patterson, *Canad. J. Chem.*, 1958, **36**, 563.
[17] W. Mackie and A. S. Perlin, *Canad. J. Chem.*, 1966, **44**, 2039.
[18] T. J. Painter, *Acta Chem. Scand.*, 1973, **27**, 2463.
[19] F. Franks, D. S. Reid, and A. Suggett, *J. Solution Chem.*, 1973, **2**, 99.

Our calculations were designed to complement and extend those of Rees and Smith in various ways. We have attempted to incorporate the effects of steric compression in a realistic way by allowing the pendant angles and the ring geometry to change in order to offset the effects of these steric interactions. We have also included entropic effects by directly estimating the free energies, as well as the energies, of the sugar conformations. Finally we have been concerned with hexoses rather than pentoses, which involves the consideration of two additional dihedral angles in specifying the orientations of rotatable side groups.

The calculations described in this paper consist of two stages. In the first, we derive appropriate geometries for the various conformations and configurations of the aldohexopyranoses in their chair forms. With the ring bond angles and dihedral angles fixed and with the substituent bond angles fixed, we then use Monte Carlo techniques to calculate the average energies and free energies of the conformers by suitable averaging over the dihedral angles of the substituents.

*Methods.*—The quality and reliability of calculations of this type are largely determined by the choice of potential function. The intramolecular conformational energy ($U$) was written as a sum of terms corresponding to non-bonded steric and dispersion terms ($U_s$), non-bonded coulombic terms ($U_c$), bond rotational (torsion) terms ($U_t$), and angle bending terms ($U_a$). $U_s$ and $U_c$ are each sums of terms over all non-bonded pairs of atoms, $U_t$ is a sum of terms over all dihedral angles and $U_a$ is a sum of terms over all valence angles. For most of the calculations the steric and dispersion terms were represented by a Lennard–Jones (6, 12) potential with the values of the constants derived by Scott and Scheraga,[20] although (6, 9) [14] and (6, exponential) [21] forms were also used in some calculations for comparison. The coulombic energy ($U_c$) was estimated as the sum of electrostatic interactions between pairs of point charges centred on the individual atoms. The charges were calculated by using the MO-LCAO method of del Re [22] and were in excellent agreement with those calculated elsewhere.[10,23] Several tests were made on the sensitivity of the calculations to the value assumed for the effective dielectric constant and good agreement between calculated and experimental geometries was obtained with values between 2 and 8. For the free energy calculations a fixed value of 4 was chosen, which is consistent with the few theoretical studies available.[24,25]

The torsional potential about each bond was assumed to be a three fold sinusoidal function of the form (i) with

$$U_i = \tau_i(1 - \cos 3\theta)/2 \qquad \text{(i)}$$

the zero of the dihedral angle ($\theta$) chosen so that the minima corresponded to the staggered conformations. The barrier heights used [23] were 2.56 kcal mol$^{-1}$ for all C–C bonds, 0.96 kcal mol$^{-1}$ for C–O bonds in hydroxy groups, and 2.4 kcal mol$^{-1}$ for other C–O bonds. (These values correspond to experimental results [26] with the contribution from non-bonded interactions removed.)

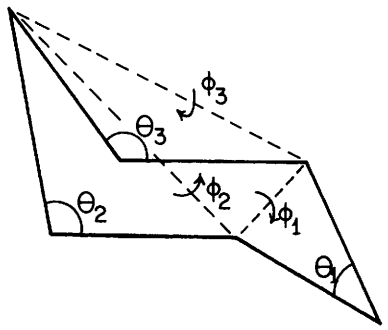
The bond angle distortion potential was assumed to be harmonic as in the usual normal mode analysis of vibrational spectra. The equilibrium bond angles and force constants [27,28] are given in Table 1.

TABLE 1

Equilibrium bond angles and force constants for the harmonic distortion potential

| Angle | Equilibrium value (°) | Force constant (kcal mol$^{-1}$ rad$^{-2}$) | Ref. |
|---|---|---|---|
| C–C–C | 112.4 | 130 | 27 |
| C–C–O | 110.0 | 172 | 27 |
| O–C–O | 110.0 | 230 | 28 |
| C–O–C | 111.5 | 158 | 28 |
| O–C–H | 109.5 | 124 | 27 |
| C–C–H | 109.5 | 96 | 27 |

Since we were concerned with the free energy differences between the stable and unstable conformers of the monosaccharides it was necessary to derive the geometries of the conformers since we could not make use of $X$-ray data for the less stable forms. Our approach was to assume the bond lengths reported by Arnott and Scott [29] and to vary all bond and dihedral angles to minimize the conformational energy. It is of course necessary to ensure that the ring remains closed, and this constraint was met by describing the ring with three bond angles and three ring distortion or ' puckering ' angles, as shown in the Figure. The ring geometry is uniquely defined by the




The ring is deformed either by changing one of the three bond angles $\theta$ or by moving a ring atom and its pendant groups, which involves changing one of the dihedral angles $\phi$

bond lengths and these six variables. In our first model only atoms directly bonded to the ring were considered. The minimization problem then has a dimensionality of

[20] R. A. Scott and H. A. Scheraga, *J. Chem. Phys.*, 1966, **45**, 2091.
[21] A. I. Kitaygorodsky, *Tetrahedron*, 1961, **14**, 230.
[22] G. del Re, *J. Chem. Soc.*, 1958, 4031.
[23] D. A. Rees and P. J. C. Smith, *J.C.S. Perkin II*, 1975, 8.
[24] G. N. Ramachandran and R. Srinivasan, *Indian J. Biochem.*, 1970, **7**, 95.
[25] G. N. Patey and J. P. Valleau. *J. Chem. Phys.*, 1975, **63**, 2334.
[26] E. B. Wilson, *Adv. Phys. Chem.*, 1959, **2**, 367.
[27] R. G. Snyder and G. Zerbi, *Spectrochim. Acta*, 1967, **23A**, 391.
[28] I. D. Blackburne, R. P. Duke, R. Jones, A. R. Katritzky, and K. A. F. Record, *J.C.S. Perkin II*, 1972, 332.
[29] S. Arnott and W. E. Scott, *J.C.S. Perkin II*, 1972, 324.

twenty-one (the six ring variables and three pendant bond angles at each of five ring carbon atoms), the remaining angles being determined from geometrical considerations and a linear dependency relation.[30] In a second model we included all atoms, which increases the dimensionality by eight [six dihedral angles and two bond angles at C(5)]. The oxygen valence angle in each hydroxy group was assumed to be tetrahedral and the bond angles involving the two hydrogen atoms attached to C(5) were assumed to be identical. The minimizations were carried out by using a simplex search.[31]

The anomeric effect was partially taken into account by using a bond length of 1.414 Å for C(1)–O(5) in the α-sugars and a bond length of 1.429 Å in the β-sugars, and assuming harmonic bond distortion to derive an energy correction of +0.049 8 kcal mol$^{-1}$ for the shortened bond in the α-ring.

With the ring geometry and the pendant angles fixed, the energy averaged over the dihedral angles of the pendant groups was calculated by using a Metropolis-type Monte Carlo method.[15] Six dihedral angles ($\phi_1$, $\phi_2$, . . . $\phi_6$) are involved and the average energy $\langle U \rangle$ is the Boltzmann average of $U$ over these six angles, *i.e.* (ii), where $Z$ is the configuration integral [see (iii)] and β is the

$$\langle U \rangle = Z^{-1} \int \ldots \int \mathrm{d}\phi_1 \ldots \mathrm{d}\phi_6 U(\sigma_1, \phi_2, \ldots \phi_6) \exp\{-\beta U(\phi_1, \ldots \phi_6)\} \quad \text{(ii)}$$

$$Z = \int \ldots \int \mathrm{d}\phi_1 \ldots \phi_6 \exp\{-\beta U(\phi_1 \ldots \phi_6)\} \quad \text{(iii)}$$

reciprocal temperature in energy units. In the Metropolis approach $U$ is averaged over successive states in a realization of a Markov chain whose unique limit distribution is proportional to the Boltzmann factor. The method has been described elsewhere.[32] The advantage of Metropolis sampling is that in order to obtain mechanical properties such as the energy it is never necessary to evaluate $Z$. However, to estimate statistical properties such as free energy or entropy one is obliged to use a more elaborate approach. To obtain abundance ratios (*e.g.* α : β ratios) one has to estimate a free energy difference quite accurately and the most convenient approach seems to be a technique originally devised by Valleau and Card,[33] which has been lucidly described.[33,34] The approach is to write the potential as a reference potential ($U_{\mathrm{ref}}$) plus a perturbation ($U'$), and to calculate the free energy difference between the reference system and the system of interest. If the free energy of the reference system is known the free energy of the system of interest is readily obtainable. We chose $U_{\mathrm{ref}} = U_{\mathrm{t}}$ and $U' = U_{\mathrm{s}} + U_{\mathrm{c}}$, since $U_{\mathrm{a}}$ is fixed in this stage of the calculation. This has the advantage that the partition function of the reference system can be calculated analytically as (iv), where $I_0$ is a zero-order Bessel function.

$$Z_{\mathrm{ref}} = (2\pi)^6 \prod_{i=1}^{6} \exp(-\tau_i/RT) I_0(\tau_i/RT) = -3.949\,5 \text{ kcal mol}^{-1} \quad \text{(iv)}$$

[30] J. B. Hendrickson, *J. Amer. Chem. Soc.*, 1961, **83**, 4537.
[31] J. A. Neelder and R. Mead, *Computer J.*, 1965, **7**, 388.
[32] See *e.g.* J. M. Hammersley and D. C. Handscomb, 'Monte Carlo Methods,' Methuen, London.

*Results and Discussion.*—Monomer geometries were derived for glucose, galactose, gulose, mannose, and idose by first ignoring all atoms not bonded directly to the ring. Bond angles obtained in this manner deviated from crystallographic values [29] by less then 2° for all angles, and in most cases by less than 1°. Those angles deviating the greatest from experimental values were C(6)C(5)O(5) and those involving C(1). The anomeric effect was clearly indicated by increases in the O(5)C(1)-O(1) and C(5)O(5)C(1) bond angles in the α-sugars. Ring

TABLE 2

Optimized co-ordinates for β-D-glucose

| Atom | $x$/Å | $y$/Å | $z$/Å |
|---|---|---|---|
| C(1) | −1.266 | 0.697 | −0.481 |
| C(2) | 0.000 | 0.000 | 0.000 |
| C(3) | 0.006 | −1.461 | −0.423 |
| C(4) | −1.290 | −2.140 | 0.000 |
| C(5) | −2.500 | −1.349 | −0.485 |
| O(5) | −2.417 | 0.000 | 0.000 |
| O(1) | −1.293 | 2.000 | 0.000 |
| O(2) | 1.141 | 0.648 | −0.552 |
| O(3) | 1.115 | −2.131 | 0.179 |
| O(4) | −1.341 | −3.454 | −0.551 |
| C(5) | −3.808 | −1.958 | −0.028 |
| H(1) | −1.280 | 0.716 | −1.580 |
| H(2) | 0.044 | 0.055 | 1.098 |
| H(3) | 0.098 | −1.516 | −1.518 |
| H(4) | −1.320 | −2.206 | 1.098 |
| H(5) | −2.495 | −1.334 | −1.586 |
| H[O(1)] | −2.091 | 2.437 | −0.304 |
| H[O(2)] | 1.143 | 1.571 | −0.288 |
| H[O(3)] | 1.931 | −1.703 | −0.088 |
| H[O(4)] | −0.581 | −3.958 | −0.249 |
| O(6) | −3.914 | −3.303 | −0.500 |
| H(a) | −3.851 | −1.950 | 1.062 |
| H(6b) | −4.639 | −1.370 | −0.418 |
| H[O(6)] | −4.744 | −3.673 | −0.202 |

TABLE 3

Optimized co-ordinates for β-D-idose

| Atom | $x$/Å | $y$/Å | $z$/Å |
|---|---|---|---|
| C(1) | −1.266 | 0.689 | −0.491 |
| C(2) | 0.000 | 0.000 | 0.000 |
| C(3) | 0.003 | −1.461 | −0.423 |
| C(4) | −1.291 | −2.144 | 0.000 |
| C(5) | −2.503 | −1.351 | −0.478 |
| O(5) | −2.418 | 0.000 | 0.000 |
| O(1) | −1.293 | 2.006 | −0.050 |
| O(2) | 0.106 | 0.085 | 1.417 |
| O(3) | 0.147 | −1.554 | −1.842 |
| O(4) | −1.339 | −2.281 | 1.419 |
| C(5) | −3.812 | −1.954 | −0.014 |
| H(1) | −1.280 | 0.682 | −1.591 |
| H(2) | 0.871 | 0.499 | −0.450 |
| H(3) | 0.851 | −1.971 | 0.057 |
| H(4) | −1.325 | −3.145 | −0.455 |
| H(5) | −2.504 | −1.340 | −1.579 |
| H[O(1)] | −2.093 | 2.433 | −0.363 |
| H[O(2)] | 0.104 | 1.007 | 1.683 |
| H[O(3)] | 0.962 | −1.124 | −2.110 |
| H[O(4)] | −0.575 | −2.780 | 1.718 |
| O(6) | −3.917 | −3.301 | −0.480 |
| H(6a) | −3.852 | −1.942 | 1.075 |
| H(6b) | −4.642 | −1.367 | −0.408 |
| H[O(6)] | −4.748 | −3.670 | −0.181 |

[33] J. P. Valleau and D. N. Caid, *J. Chem. Phys.*, 1972, **57**, 5457.
[34] G. M. Torrie and J. P. Valleau, *Chem. Phys. Letters*, 1974, **28**, 578.

torsional angles agreed well with the Arnott and Scott averages.[29]

Calculations involving all atoms were carried out for all sixteen hexose monomers with slightly better agreement (to within 1.5° of the crystallographic values). The final co-ordinates calculated for β-glucose are given in Table 2 and those for β-idose are given in Table 3, for comparison.

For both models the more crowded isomers showed a definite flattening of the ring. Replacing an equatorial substituent by an axial one gave rise to an increase in pendant angles, in agreement with experimental evidence. As both models give similar results, further discussion will centre on the full atom treatment only.

The average energies, free energies, and entropies found from the Monte Carlo calculation are given in Table 4.

TABLE 4 †

| Hexose | Ring conformation | Internal energy | Entropy | Free energy |
|---|---|---|---|---|
| α-Glucose | *C*1 | 3.07 | 14.73 | −1.38 |
| | 1*C* | 3.76 | 13.79 | −0.41 |
| β-Glucose | *C*1 | 3.17 | 15.42 | −1.51 |
| | 1*C* | 4.60 | 12.17 | 0.93 |
| α-Galactose | *C*1 | 3.01 | 14.21 | −1.28 |
| | 1*C* | 4.09 | 13.62 | −0 02 |
| β-Galactose | *C*1 | 3.29 | 15.02 | −1.25 |
| | 1*C* | 5.00 | 12.07 | 1.38 |
| α-Mannose | *C*1 | 2.96 | 14.60 | −1.45 |
| | 1*C* | 3.26 | 14.15 | −1.01 |
| β-Mannose | *C*1 | 3.62 | 15.15 | −0.94 |
| | 1*C* | 5.03 | 13.28 | 1.18 |
| α-Gulose | *C*1 | 3.34 | 13.90 | −0.86 |
| | 1*C* | 4.34 | 14.19 | −0.05 |
| β-Gulose | *C*1 | 2.76 | 14.72 | −1.71 |
| | 1*C* | 5.01 | 12.53 | 1.23 |
| α-Idose | *C*1 | 3.29 | 13.58 | −0.81 |
| | 1*C* | 3.88 | 14.23 | −0.41 |
| β-Idose | *C*1 | 3.30 | 14.37 | −1.04 |
| | 1*C* | 4.58 | 12.38 | 0.85 |
| α-Allose | *C*1 | 4.04 | 14.27 | −0.26 |
| | 1*C* | 4.65 | 13.71 | 0.51 |
| β-Allose | *C*1 | 3.29 | 15.14 | −1.28 |
| | 1*C* | 6.04 | 11.96 | 2.43 |
| α-Altrose | *C*1 | 3.27 | 14.59 | −1.13 |
| | 1*C* | 3.57 | 14.94 | −0.94 |
| β-Altrose | *C*1 | 3.30 | 15.15 | −1.27 |
| | 1*C* | 4.49 | 12.99 | 0.57 |
| α-Talose | *C*1 | 3.44 | 13.41 | −0.61 |
| | 1*C* | 4.43 | 13.78 | 0.27 |
| β-Talose | *C*1 | 4.30 | 14.03 | 0.05 |
| | 1*C* | 5.67 | 11.52 | 2.19 |

† Energy and free energy in kcal $mol^{-1}$ and entropy in cal $mol^{-1}$ $K^{-1}$. Standard deviations are of the order of 0.03, 0.15, and 0.05, respectively, in the above units.

As free rotation would correspond to an entropy of 21.9 cal $mol^{-1}$ $K^{-1}$ one can conclude that there is a large degree of rotational freedom for the substituents. β-Sugars invariably have a higher entropy than the corresponding α-sugars, and increasing the number of axial groups leads to the expected lowering of the entropy.

The proportion of each D-isomer in the 1*C* chair form is given in Table 5. The calculations predict that the 1*C* form is most important for α-mannose, α-idose, and α-altrose, which is also suggested by n.m.r. evidence,[35] but the overall amounts of the 1*C* forms for the α-sugars are almost certainly too large. (It is interesting that assumption of the *C*1 chair form leads to poor agreement between calculated and experimental optical rotations for these three sugars.[36]) As was to be expected the amount of 1*C* chair predicted for the β-sugars was low, with even sterically hindered β-idose having only 4% of the alternative chair. Table 5 also includes the calculated α : β ratios and their experimental values. The free energy estimates are generally satisfactory but tend to overestimate the abundance of α-sugars, particularly for D-galactose. However the general trends in the entire series of pyranoses are encouraging, even for the sterically hindered cases of idose and talose. In comparison Angyal,[8,9] while arriving at excellent α : β ratios for the sterically unhindered hexoses, did not predict the correct direction of the idose ratio. It is important to remember that Angyal's free energy estimates were chosen so as to reproduce correctly the α : β ratios for glucose, mannose, and allose. In the present work, however, no attempt has been made to ' build in ' these ratios, and all parameters are generally accepted ones taken from the literature. The reproduction of α : β ratios for the entire hexose series is an indication of the validity of this general model, and suggests that it will serve as a convenient basis for polysaccharide calculations.

TABLE 5

| Hexose | % in 1*C* form | | % in α form † | | |
|---|---|---|---|---|---|
| | α-Hexose | β-Hexose | calc. (a) | calc. (b) | exp. |
| Glucose | 17 | 1.6 | 47 | 33 | 37 |
| Mannose | 33 | 2.8 | 79 | 64 | 67 |
| Galactose | 10 | 1.2 | 54 | 41 | 27 |
| Gulose | 21 | 0.7 | 23 | 15 | 21 |
| Idose | 34 | 4.1 | 50 | 39 | 40 |
| Allose | 22 | 0.2 | 19 | 17 | 20 |
| Altrose | 42 | 4.4 | 57 | 50 | 40 |
| Talose | 19 | 2.8 | 76 | 69 | 58 |

† Values (a) have no solvation correction; values (b) include the solvation correction.

The free energies are not entirely satisfactory however. The α : β ratio for galactose is too high and the prediction of β-gulose as a particularly stable isomer is surprising. (Rao [10–13] found β-gulose to be the next most stable isomer after β-glucose.) If the pyranose rings were made less flexible, then the energy differences might conform more closely to those of Angyal, but two arguments rule out such an approach. First, Angyal's results are not to be interpreted as the exact experimental values and so agreement with them would not necessarily increase our confidence in the present model. Indeed his results are certainly inaccurate in the case of idose. Secondly, to make any parameter adjustable would defeat the present purpose of trying to apply general principles to an energy calculation for the pyranoses. The bond bending force constants used here are experimentally observed quantities and there is no justification for changing them.

The observation that whereas glucose is 36.6% α-form

[35] M. Rudham and D. F. Shaw, *J. Chem. Soc.*, 1965, 52.
[36] J. H. Brewster, *J. Amer. Chem. Soc.*, 1959, **81**, 5475, 5483.

in aqueous solution [36] in dry pyridine it is 43% α-anomer [37] gives a possible explanation for the discrepancy in the present results. Our calculations ignore solvent effects, although it is known that hexoses form strong hydrogen bonds with aqueous solvents. Kabayama and Patterson [16] have suggested that the pyranose ring fits particularly well into the ice-like solvent lattice of aqueous solutions. Hydrogen bonding to equatorial groups stabilizes this lattice about the ring, whereas the presence of axial groups disrupts the structuring of water above and below the ring. These suggestions are supported by the work of Mackie and Perlin [17] and by Franks *et al.*,[19] who found that water forms hydrogen bonds to methyl α- and β-glycosides in different ways. Both anomers of glucose and β-galactose form particularly stable hydrogen bonds.

A simple model [16] for such solutions, is to assign an energy value of −0.27 kcal mol$^{-1}$ to each hydrogen bond. Further, the number of hydrogen bonds formed is equal to the number of equatorial hydroxy groups present in the *C*1 chair. No hydrogen bonds are assigned to the 1*C* chair of the D-isomers, as the axial $CH_2$·OH group would be expected to disrupt any structuring of a water lattice.

The values of the α : β ratios corrected in this way are also given in Table 5. Agreement with the experimental results is good in every case except for galactose, in which the α : β ratio is badly overestimated. This may be due to the extra solvation effects observed for β-galactose.[19] It is especially pleasing that the calculated α : β ratio for glucose, after this simple correction for solvation, agrees well with the experimental value in aqueous solution; before the solvation correction it agreed well with the experimental value in dry pyridine.

There are a number of estimates of free energy differences between monosaccharides from experimental work on certain enzyme-catalysed epimerizations.[38] We have compared our estimates of the free energy differences with a number of these experimental estimates, and found the agreement of both direction and magnitude of the equilibria to be generally satisfactory. As an example, we consider the epimerization of α-D-glucose to α-D-galactose. For the unsolvated free sugars we estimate the former to be 0.1 kcal mol$^{-1}$ more stable than the latter, and this free energy difference is increased to 0.37 kcal mol$^{-1}$ by including the solvation correction. Maxwell [39] has studied the epimerization of UDP-α-D-galactose to UDP-α-D-glucose and estimates the difference to be 0.66 kcal mol$^{-1}$. The corresponding difference for the 1-phosphates is estimated to be 0.7 kcal mol$^{-1}$.[40] Considering the bulk of the substituents, the agreement with experiment is reasonable.

As a test of the suitability of the optimized monomer co-ordinates for use in calculations of the random coil dimensions of polysaccharides, we have estimated the characteristic ratios of cellulose and amylose. To simplify the calculations the monomer geometries were calculated with inclusion of all substituents but, in the calculation of the characteristic ratios of the polymers, all hydroxy groups were approximated by single oxygen atoms. [The rotations about C(5)–C(6) were included in the calculations.] The method used is standard.[41,42] The calculated values of 7.7 and 45 for amylose and cellulose are to be compared with the experimental values [4,5] of 6.7 and 35. The agreement is good in view of the approximations inherent in such a calculation, and represents more satisfactory results than most prior work,[42,43] in which ideal or average [44] co-ordinates were used.

*Conclusions.*—The primary objects of this work have been to explore the feasibility of (*a*) calculating monomer geometries and (*b*) estimating free energy differences between epimers. In the case of monomer geometries we have chosen to simplify the calculation by assuming known fixed bond lengths and to find optimum values of the valence and dihedral angles. In general our results are in excellent agreement with crystallographic values. Using these geometries we have estimated free energies of each of the hexose monomers by extensions of a Metropolis-type Monte Carlo method. Our results are in reasonable overall agreement with experiment but the agreement is much improved if one includes a simple solvation correction. Indeed we believe that our results strongly support the importance of solvation effects in aqueous solutions of monosaccharides.

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