A model for the graphical display of pseudorotation in small rings

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An improved mathematical description of pseudorotation in five- and six-membered rings is presented. This procedure allows the position of the ring atoms to be computed at all points on the pseudorotational pathway without significant geometrical distortion, bond lengths remaining constant to within ± 2A° and endocyclic valence angles to within ± 2°. The routine has been incorporated into an interactive molecular modeling system, allowing real-time display of the pseudorotation process in molecules of biological interest and calculation of associated conformational energies and NMR coupling constants.

INTRODUCTION

Saturated five-membered ring systems — such as the amino acid proline, the furanose ring of nucleosides and the cyclopentane ring of prostaglandins (Figure 1) — are commonly found in biologically important molecules. These rings often possess considerable conformational flexibility due to the low energy barriers to interconversion between different ring conformations. The furanose ring system in nucleosides and nucleotides is known to exist in two common conformational states in both solid state and solution,[1,2] and four distinct conformations have been observed in oligonucleotides.[3] Similarly, the proline ring system is found to exist in two major conformational classes.[4] This conformational variability can markedly affect the spatial relationship between the substituent groups. As compounds containing saturated five-membered rings are becoming increasingly important as pharmaceutical agents, an accurate model of the interconversion processes between these different conformations would be valuable in studying structure-activity relationships in these molecules.

MATHEMATICAL MODEL

The motion of ring atoms in the interconversion of related conformations in five- and six-membered rings, such as the C2'-endo and C3'-endo conformers in furanoses[2] and twist-boat forms of cyclohexane, can be described by the concept of pseudorotation. For cycloalkanes, the displacement Z_j of an individual ring atom j from a model reference frame — the regular flat polygon resulting from the projection of the ring onto the least-squares plane — is given by a harmonic function[5]:

$$Z_j = Z_m \cos (\phi + 4\pi j/n) \qquad (j = 0, n-1) \qquad (1)$$

where n = number of ring atoms
Z_m = maximum out-of-plane displacement
ϕ = pseudorotational phase angle

In this function, the phase angle ϕ locates the position of a conformer on the pseudorotation pathway relative to a chosen reference (note that the definition of axes and origins can cause problems — we follow a convention that if the ring numbering is viewed clockwise, then positive Z should come toward the viewer. The definition of the phase angle is based on torsion angles[6] and is related to that based on Z-displacements by a phase shift of $\pi/2$). However, saturated ring systems normally show a significant ring pucker — that is, a large value of Z_m relative to standard bond lengths — and simple application of Equation 1 will lead to serious geometrical distortion of both bond lengths and angles during different phases of the pathway.[7] We have therefore developed an improved mathematical description of pseudorotation that allows for the additional motion of the atoms in the plane of the reference frame and hence maintains the geometrical integrity of the ring. The polar atomic coordinates R_j and θ_j, which describe the in-plane movement of the atoms about the origin of motion in regular systems (e.g., cycloalkanes) both
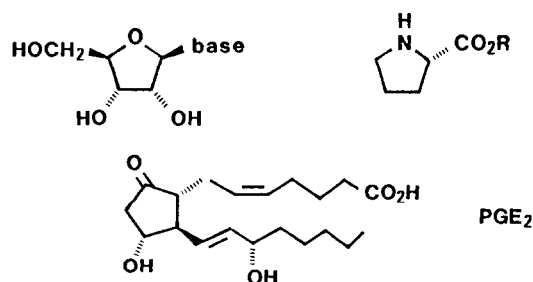

Figure 1. Biologically important five-membered rings: the nucleoside ribose ring, proline and prostaglandin PGE-2
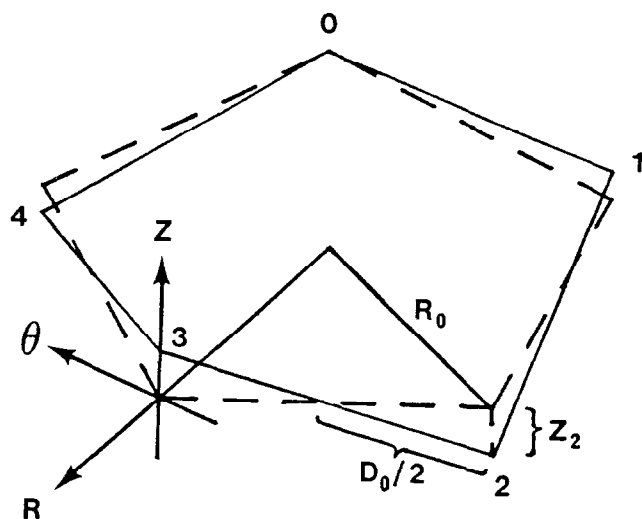
Figure 2. Projection of cyclopentane onto the least-squares plane, showing the direction of the in-plane (θ, R) and out-of-plane (Z) atom motions. The pseudorotation phase angle, with respect to torsions, is set to zero

along the polygon radius vectors and at right angles to them (Figure 2), are given by:

$$R_j = R_o(1 + A_n\cos(2\phi + 8\pi j/n)) \tag{2}$$

$$\theta_j = (2\pi/n)(j + B_n\sin(2\phi + 8\pi j/n)) \tag{3}$$

where R_o is the radius of the regular polygon and A_n and B_n are constants that can be adjusted to minimize the change in ring geometry during pseudorotation. These additional formulas show a frequency twice that for displacements above and below the ring plane (Equation 1). The radius R_o can be calculated from Z_m and the internuclear distance D_o (Figure 2) by the expression

$$R_o = \{(D_o^2 - 2Z_m^2\sin^2(2\pi/n))/2(1 - \cos(2\pi/n))\}^{\frac{1}{2}}$$

$$(n = 5,6) \tag{4}$$

For nonregular heterocyclic systems (see below) D_o can be replaced by the averaged bondlength between the ring atoms. Z_m can be determined from the sums-of-squares of the observed Z-displacements for any phase angle[5]:

$$Z_m = \left\{ (2/n)\sum_i Z_i^2 \right\}^{\frac{1}{2}} \tag{5}$$

The distance D_{ij} between the ith and jth atom of a nonplanar polygon (Figure 2) is given by:

$$D_{ij}^2 = R_i^2 + R_j^2 - 2R_iR_j\cos(\theta_i - \theta_j) + (Z_i - Z_j)^2 \tag{6}$$

and by substitution into Equation 6 with Equations 1–3, it can be shown that the distance D_{ij} remains constant if

$$2R_o^2 \left\{ A_n\cos(4\pi(i-j)/n)(1 - \cos(2\pi(i-j)/n)) \right.$$
$$\left. + B_n\sin(4\pi(i-j)/n)(\sin(2\pi(i-j)/n)) \right\}$$
$$= Z_o^2\sin^2(2\pi(i-j)/n) \tag{7}$$

There is no clear experimental reason for assuming that endocyclic bondlengths do not remain essentially constant (i.e., within $\pm$ 0.01 Å) during pseudorotation, although we note that an alternative description of the

pseudorotational process in furanoses explicitly requires that bondlengths vary periodically with the phase angle.[8] Substitution into Equation 7 for constant 1–2 distances in five-membered rings gives

$$B_5 - A_5 = Z_m^2/(4R_o^2\cos(2\pi/5)) \tag{8}$$

Apparently, any linear combination of A_5 and B_5 may be taken. Operationally it was found possible to set B equal to zero, allowing motion only along the radius vectors. Hence

$$A_5 = -0.8090 \, Z_m^2/R_o^2 \qquad B_5 = 0 \tag{9}$$

The center of motion for five-membered rings is not exactly at the centroid, and small corrections must be applied from the centroid in the least-squares plane of:

$(A_5R_o\cos(2\phi))/2$ along the 1st radius vector

$(A_5R_o\sin(2\phi))/2$ perpendicular to the 1st radius vector, on the same side as the 2nd

For six-membered and larger rings both constant 1–2 and 1–3 distances may be achieved, giving pairs of simultaneous equations that allow us to solve for both A and B

$$A_6 = -3 \, Z_m^2/(4R_o^2) \qquad B_6 = Z_m^2/(4R_o^2) \tag{10}$$

For any given molecular structure — for example, an X-ray structure — the phase angle ϕ and the parameters Z_m and R_o can be calculated uniquely. By application of Equations 1–3 a set of dummy ring atom coordinates for the starting conformation are calculated, and the movement of these dummy atoms relative to the least-squares plane for a given increment in the phase angle ϕ may be determined. The components of the nett change in these displacements are then transformed into Cartesian space and added to the original true coordinates to give a new set of ring atom coordinates. We found that no modification of Equations 1–3 was necessary to account for the motion of heterocyclic systems. In the pseudorotational cycle of five-membered rings, ring bondlengths are found to remain constant to within 0.01 Å, even in nonregular heterocyclic systems, and endocyclic bond angles vary by $\pm$ 2° in a similar manner to known systems.[9,10] For example, the C4'–O4'–C1' valence angle in furanose rings was found to vary from a maximum tetrahedral value of 109°, at phase angles of 0° and 180°, to a minimum of 105° at phase angles of 90° and 270°; the valence angles at C2' and C3' conversely increased from 102° to 104° at the same points. In six-membered rings, using the interconversion of twist-boat forms of cyclohexane as a model, ring bondlengths were found to vary by up to $\pm$ 0.02 Å, while valence angles were constant to within 1°.

If the exocyclic valence angles made by the substituents at their point of attachment to the ring are assumed to remain constant, the motion of the attached fragments can be calculated analytically. This assumption is approximate, since angle deformation of the order $\pm$ 2° involves only a low energy penalty,[9] but it provides a useful model for gross atomic movements.

Table 1. Additional torsional parameters used in rigid-rotor force-field calculations

C2 = C(sp2) C3 = C(sp3) N2 = N(sp2)
O2 = O(sp2) O3 = O(sp3)
$E_\theta = K_\theta *(1 - a*\cos(n\theta))$

Torsion	K_θ	a	n
H-C3-C3-H	0.15	1	3
C3-C3-C3-H	0.18	1	3
C3-C3-C3-C3	0.18	1	3
H-C3-C3-O3	0.22	1	3
C3-C3-C3-O3	0.15	1	3
H-C3-O3-C3	0.36	1	3
C3-C3-O3-C3	0.25	1	3
O3-C3-C3-O3	0.25	1	2
O3-C3-C3-N2	0.12	1	2
H-C3-C3-N2	0.15	1	3
C3-C3-C3-N2	0.12	1	3
H-C3-N2-C3	0.12	1	3
C3-C3-N2-C3	0.12	1	3
H-C3-C2=O2	0.10	-1	3
C3-C3-C2=O2	0.10	-1	3

Other torsional and nonbonded parameters may be found in Ref 12

APPLICATIONS

Routines based on Equations 1-3 have been incorporated into a molecular modeling system to permit interactive display of five- and six-membered rings through the pseudorotation pathway. Real-time calculation and graphical display of molecular mechanics energies and of proton-proton coupling constants have been implemented. The molecular mechanics energies are calculated using a "rigid-rotor" forcefield, comprising nonbonded, torsional and electrostatic potential functions.[11,12] Only amide groups have been specifically allotted partial electrostatic charges, as interactions between adjacent polar groups have been allowed for in the torsional potentials (Table 1) (for example, two-fold torsional potentials have been used to model the gauche conformational preferences of sp3-hybridized O-C-C-O and related systems[13]). As the nett energy variation arising from endocyclic bond angle and bond-length deformation in our model will not amount to more than 0.2 kcal/mole for all phases of the pseudorotation cycle, assuming deviations from standard tetrahedral geometries,[11] no account was made of these components in the energy calculations. Displays of the pseudorotational processes in cyclopentane and twist-boat cyclohexane, shown in Color Plates 1 and 2, illustrate interconversion barrier heights of essentially zero for cyclopentane and about 1.0 kcal/mole for cyclohexane. These values are similar to those determined by other computational procedures.[14] NMR coupling constants are calculated using a modified Karplus-type function[15] and may be displayed for either specified pairs of vicinal protons or as a root-mean-square (rms) difference curve from experimentally determined values.

N-Acetyl-L-Proline-N'-Methylamide

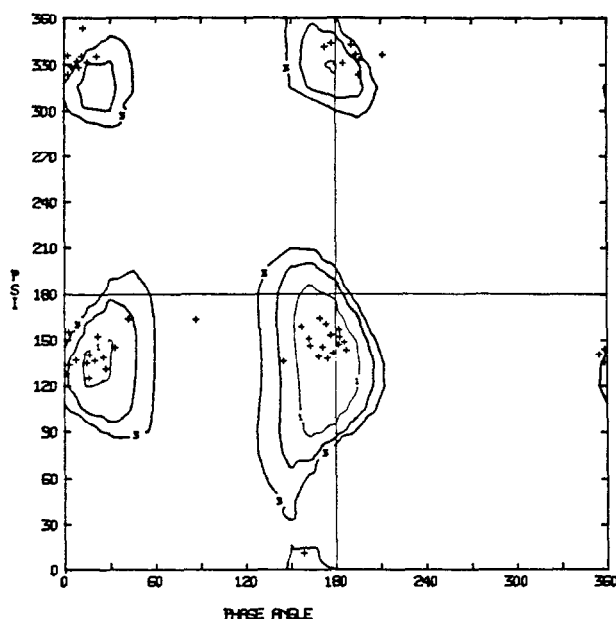Real map contoured every 1 Kcal per mole

Figure 3. Conformational energy map of N-acetyl-L-proline-N'-methylamide as a function of the phase angle ϕ and the backbone peptide torsion ψ. The crosses (+) represent conformations of N-alcyl-L-proline-N'-alkyl-amides found in the Cambridge Crystallographic Database

The addition of substituents can severely restrict the pseudorotational process. Color Plate 3 shows the calculated energy curve for the prostaglandin PGE-2, together with a display of the rms difference between the calculated and observed ring vicinal coupling constants for different points on the pseudorotational cycle. This shows a good correspondence between the calculated energy minimum and the minimum rms for the coupling constants. Similar curves have been determined for proline,[12] and our experience has shown that if we possess three or more experimental coupling constants and an rms of 1.0 Hz or better is achieved, then the molecule almost certainly exists in a single unique conformation.

As a further refinement, an exocyclic torsion angle can be driven independently of the pseudorotation process. This permits the calculation of two-dimensional energy plots, with the pseudorotational phase angle and the chosen torsion angle as axes. Plots for N-acetyl-L-proline-N'-methylamide and 2'-deoxycytidine are shown in Figures 3 and 4, together with the observed distribution of conformers found in the Cambridge Crystallographic Database.[16] The barriers to interconversion of the C-2'-endo and C-3'-endo ribose conformations are calculated to be 2.5 kcal/mole (Color Plate 4), and slightly lower (2 kcal/mole) in the 2'-deoxyribose series (Figure 4). These figures are for fixed glycosidic torsion angles in both purine and pyrimidine nucleosides (X = -135°; glycoside anti conformation) and are based on ring geometries obtained from high resolution X-ray studies (i.e., they have not been subjected to energy

2'-Deoxycytidine

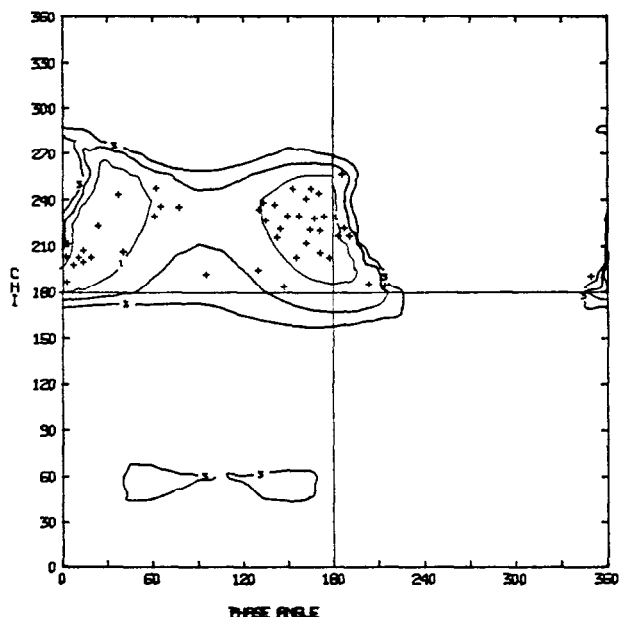Real map contoured every 1 Kcal per mole

Figure 4. Conformational energy map of 2'-deoxycytidine as a function of the phase angle φ and the glycosidic torsion X. The crosses (+) represent conformations of 2'-deoxy pyrimidine nucleosides found in the Cambridge Crystallographic Database

minimization procedures). The energy difference between the C2'-endo and C3'-endo conformers amounts to no more than 0.5 kcal/mole, in favor of the C3'-endo form, in any series. These differences are small, and must be viewed carefully when considering the approximations inherent in trying to apply molecular mechanics force fields to the complex dynamic processes present in riboses.[17] However, the values are close to those predicted in alternative models of the pseudorotation,[13,17,18] and the good correspondence between the calculated energy minima shown in Figures 3 and 4 and the distribution of experimental X-ray conformers leads us to believe that the method can be used to confidently model the energetic processes in these chemical systems.

ACKNOWLEDGEMENTS

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