Acknowledgment. This work was supported by the National Science Foundation, CHE-9014941.

Supplementary Material Available: Complete specifications of the geometries of the reactants, TS, and product from the 3-21G optimizations in Z-matrix format and the potential function parameters (3 pages). Ordering information is given on any current masthead page.

Evidence for a Dominant Suprafacial-Inversion Pathway in the Thermal Unimolecular Vinylcyclopropane to Cyclopentene 1,3-Sigmatropic Shift

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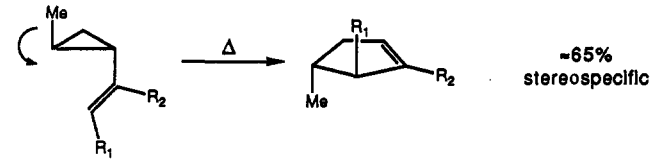
All previous studies of the thermal, unimolecular isomerization of vinylcyclopropane to cyclopentene[1] demonstrated dominant suprafacial-inversion stereochemistry in this 1,3-sigmatropic shift, but relied on *trans*-2-methyl substitution at the migrating carbon to reveal the pathway[2,3] (Scheme I).

Concern that steric effects may be responsible for the stereoselectivity by forcing outward rotation of the *trans*-methyl group as the C-1,C-2 bond breaks, which might present the back side of the migrating carbon to the end of the allylic moiety for a least motion closure,[4] prompted examination of the pyrolysis of *r*-1,*t*-2,*t*-3-2,3-dideuterio-1-[(*Z*)-1′-*tert*-butyl-2′-deuteriovinyl]-cyclopropane (**1**). A complication is the geometric isomerization of starting vinylcyclopropane, which in the parent case occurs ca. 20 times faster than the 1,3-shift[1,2] and gives a nearly statistical distribution of diastereomers at all three ring centers.[5]

Vinylcyclopropane **1** was chosen for study in the anticipation that the *tert*-butyl group would retard formation upon pyrolysis of a transoid allylic species which could only reclose to starting material and compromise its stereochemistry. Indeed, it was found that, upon pyrolysis at 290 °C, **1** (for synthesis, see supplementary material) undergoes formation of a random mixture of cyclopropane ring diastereomers[5] only 4.5 times faster than 1,3-shift.

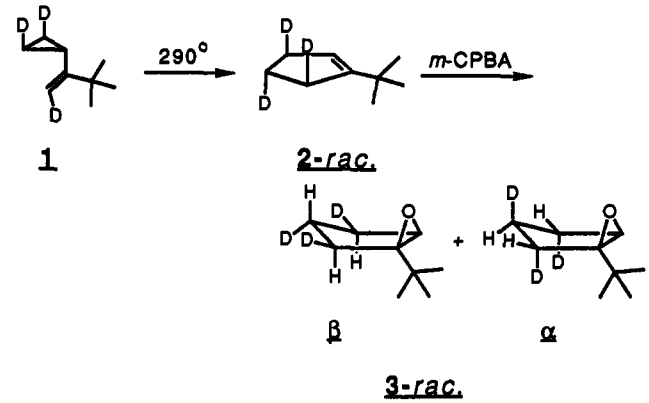
Fortunately, all of the ring protons of the epoxide of protio 1-*tert*-butylcyclopentene were found to be separately visible in the 500-MHz ^{1}H NMR spectrum and are assignable on the basis of a low-energy boat conformation.[6] Molecular mechanics found the same low-energy conformation of the product epoxide,[7] and the coupling constant calculation protocol of Haasnoot[8] produced coupling constants similar to those observed. Of particular note is that there are two trans coupling systems on the ring, with one (α) having J_{trans} = 0 Hz, which corresponds to the endo C-2 and endo C-4 protons interacting with the exo C-3 proton, and the other (β) corresponding to exo C-2 and exo C-4 protons interacting with the endo C-3 proton with J_{trans} = 9 Hz. The cis coupling constants are not inconsequential (J_{cis} = 8–9 Hz) and lead to doubling of the C-2 and C-4 protons and tripling of the C-3 protons, all in addition to the doubling by geminal coupling (J_{gem} = 12–14 Hz).

Scheme I[a]



[a] R_1 = Me, R_2 = H (ref ^ R_1 = D, R_2 = *t*-Bu (ref 3).

Observation of the ^{1}H NMR spectra (500 MHz; unlocked, deuterium decoupled) of the epoxidized, GC-purified cyclopentene product resulting from the rearrangement of **1** over varying pyrolysis times showed that the rearrangement is not random. Samples of **1** (90% C-2 and C-3 deuterium incorporation trans to C-1 substituent; >98% deuterium incorporation cis to *tert*-butyl) were pyrolyzed to 60%, 6.4%, and 4.5% conversion to **2**. The



shorter the pyrolysis time, the more closely the ^{1}H NMR spectrum of the product epoxide resembles that predicted for a purely suprafacial inversion process, and the less it resembles that predicted for a random mixture. In particular, the multiplets associated with the exo C-2 and C-4 and endo C-3 protons from the *sr*, *ar*, and *ai* components in the mixture decrease in intensity, leaving the singlets predicted for α, which is the *si* product.[9]

It is not possible to calculate the stereospecificity in the 1,3-shift to high accuracy, but it appears to be in excess of 85%. Regardless of the exact value, it is clear that the 55–80% suprafacial-inversion stereochemistry observed with 2-*trans*-alkyl-substituted vinylcyclopropanes is not a result of steric effects.

Demonstration of suprafacial-inversion stereochemistry is consistent with concert in the rearrangement with a competing, entropically more favorable, biradical pathway which randomizes the stereochemistry of starting material. The stereochemistry is also consistent with disrotatory ring opening followed by rapid (relative to bond rotation) closure of the biradical resulting from inward rotation of the vinyl group; the biradical resulting from outward rotation of the vinyl group might be responsible for the loss of stereointegrity of the starting material. The origin of the disrotatory ring opening stereomode is not obvious. The observation of a substantial *normal* deuterium kinetic isotope effect at the exomethylene carbon in the rearrangement of vinylcyclopropane itself[10] and of 2-methyl-1-vinylcyclopropanes,[3] which is

(1) (a) Neureiter, N. P. *J. Org. Chem.* **1959**, *24*, 2044. (b) Marshall, D. C.; Frey, H. M. *J. Chem. Soc.* **1962**, 3981.

(2) Andrews, G. D.; Baldwin, J. E. *J. Am. Chem. Soc.* **1976**, *98*, 6705. **Note Added in Proof:** Baldwin and Ghatlia (Baldwin, J. E.; Ghatlia, N. D. *J. Am. Chem. Soc.* **1991**, *113*, 6273) found similar behavior for the case with R_1 = D and R_2 = H.

(3) Gajewski, J. J.; Squiciarini, M. P. *J. Am. Chem. Soc.* **1989**, *111*, 6717.

(4) W. von E. Doering, suggestion made at the 1988 Reaction Mechanisms Conference, University of Pittsburgh.

(5) Willcott, M. R., III; Cargle, V. H. *J. Am. Chem. Soc.* **1969**, *91*, 4310. A doublet superimposed on a doublet of doublets is observed for anti ^{1}H(s); this spectrum does not change with pyrolysis time.

(6) (a) Lafferty, W. J. *J. Mol. Spectrosc.* **1970**, *36*, 84. (b) Steyn, R.; Sable, H. Z. *Tetrahedron* **1971**, *27*, 4429.

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(8) Haasnoot, C. A. G.; deLeeuw, F. A. A. M.; Altona, C. *Tetrahedron* **1980**, *36*, 2783.

(9) One epoxide of the suprafacial-inversion product from **1** should have only endo C-2 and endo C-4 protons with an exo C-3 proton for the α coupling system (J_{trans} = 0 Hz), and since there should be no cis protons in the *si* product (and no J_{gem} regardless of stereochemistry), only three ring singlets should be observed for this epoxide. The other epoxide has protons only at exo C-2, exo C-4, and endo C-3, so the β system doublet–doublet–triplet pattern, respectively, is expected.

(10) Chickos, J. *Abstracts of Papers*, 187th National Meeting of the American Chemical Society, St. Louis, MO; American Chemical Society: Washington, DC, 1984; ORGN 228.

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most reasonably attributed to a rotational isotope effect,[11] is consistent with concert. For the isotope effect to be consistent with the stereospecific biradical pathway, closure to the five-membered ring must be rate-determining. However, it is hard to reconcile the demand of slow closure to a five-membered ring relative to closure to a three-membered ring with the demand of rapid closure to a five-membered ring relative to bond rotation in the diradical. Thus, the 1,3-shift would appear to be concerted.

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Supplementary Material Available: Experimental details for the synthesis of **1** and NMR spectra of the methylene protons of the undeuterated epoxide, the deuterated epoxide from the three different reaction times, and simulations of mixtures (9 pages). Ordering information is given on any current masthead page.

(11) Dai, S. H.; Dolbier, J. *J. Am. Chem. Soc.* **1970**, *92*, 1774; **1972**, *94*, 3946. Crawford, R. J.; Chang, M. H. *Tetrahedron* **1982**, *38*, 837. Gajewski, J. J.; Benner, C. W.; Stahly, B. N.; Hall, R. F.; Sato, R. I. *Tetrahedron* **1982**, *38*, 853.

Atomic Structure of the Rapamycin Human Immunophilin FKBP-12 Complex

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Complexes of immunophilins with immunosuppressive drugs interfere with a variety of signal transduction pathways in the cytoplasm of the cell.[1-3] Rapamycin[4] (**1**) is a high affinity ligand (K_d = 0.2 nM)[2] to the immunophilin FKBP-12[5-7] and appears to be a general and potent antiproliferative agent.[1] The pleiotropic actions of rapamycin on growth factor receptor signaling pathways have elevated this compound to a high status as a probe of signaling mechanisms. Although the precise details have yet to be elucidated, the *complex* of human FKBP-12 and rapamycin has been shown by genetic methods to function as the inhibitory agent.[8] Herein we report the three-dimensional structure of the complex of human FKBP-12 and rapamycin, determined to 1.7-Å resolution by X-ray crystallographic techniques.[9] This structure provides a framework to interpret the effects of structural perturbation of either rapamycin or human FKBP-12 on signal transduction pathways.

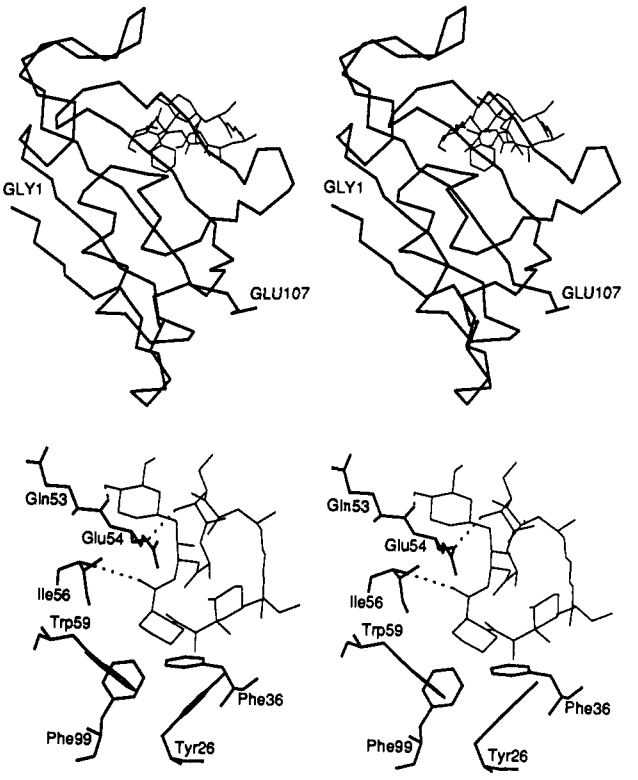
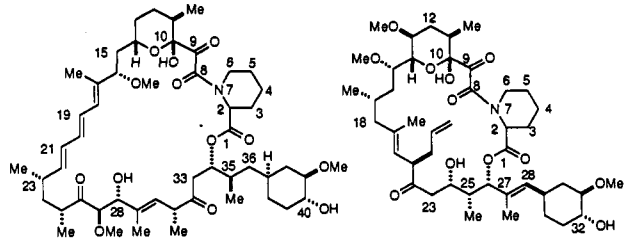


Figure 1. (a, top) A stereoview of the α-carbon tracing of FKBP-12 and rapamycin. The N- and C-terminal α-carbons are labeled. (b, bottom) A stereodrawing of the binding pocket showing all of the bound rapamycin molecule and selected FKBP-12 residues.



1 - Rapamycin 2 - FK506

As shown in Figure 1, the protein component of the FKBP-12/rapamycin complex forms a five-stranded antiparallel β-sheet

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(1) Schreiber, S. L. *Science* **1991**, *251*, 283.
(2) Bierer, B. E.; Matila, P. S.; Standaert, R. F.; Herzenberg, L. A.; Burakoff, S. J.; Crabtree, G.; Schreiber, S. L. *Proc. Natl. Acad. Sci. U.S.A.* **1990**, *87*, 9231-9235.
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(4) Findlay, J. A.; Radics, L. *Can. J. Chem.* **1980**, *58*, 579.
(5) The family of proteins that bind to FK506 and rapamycin have been collectively named FKBP's (FK506 binding proteins), with a suffix designating the approximate molecular weight. Human FKBP-12, the most abundant FKBP found in the cytoplasm, is a 12 kD protein of 107 amino acids that binds FK506 and rapamycin and catalyzes the cis-trans isomerization of peptidyl-prolyl amide bonds in peptide substrates.
(6) Harding, M. W.; Galat, A.; Uehling, D. E.; Schreiber, S. L. *Nature* **1989**, *341*, 758.
(7) Siekierka, J. J.; Hung, S. H. Y.; Poe, M.; Lin, C. S.; Sigal, N. H. *Nature* **1989**, *341*, 755.
(8) Koltin, Y.; Faucette, L.; Bergsma, D. J.; Levy, M. A.; Cafferkey, R.; Koser, P. L.; Johnson, R. K.; Livi, G. P. *Mol. Cell. Biol.* **1991**, *11*, 1718.

(9) Crystals of the FKBP-12/rapamycin complex were grown from solutions containing 10 mg/mL of protein complex, 300 mM ammonium sulfate, and 100 mM phosphate at pH 6.0 using the hanging drop method at room temperature. The space group is $P2_12_12_1$ with a = 45.42 Å, b = 49.16 Å, c = 54.74 Å, and one molecule in the asymmetric unit. Data were measured using a SanDiego Multiwire Systems Mark II detector and a rotating anode source to 1.7-Å resolution. A total of 81 484 reflections were measured (12 991 unique, 93% complete, R_{sym} = 0.056, 10 633 with $F \geq 3\sigma$) from two crystals. The structure was solved using the molecular replacement method with a search model composed of the protein component of the FKBP-12/FK506 complex and the MERLOT program system.[10] The structure was refined with X-PLOR[11] using least-squares minimization by conjugate gradients where the stereochemical restraints used in ligand refinement were restricted to terms for bond lengths, bond angles, and improper dihedral angles (for planar sp^2 carbons and chiral centers). The conformation of bound rapamycin was determined unambiguously from well-defined electron density in $2F_o-F_c$ maps. The R factor for the current model, including FKBP-12, rapamycin, and 85 water molecules, is 0.165. All main chain atoms, all buried side-chain atoms, and all ligand atoms are well-defined in the final $2F_o - F_c$ electron density map. The root-mean-square deviations of bond lengths and bond angles from their ideal values are 0.01 Å and 2.8°, respectively.
(10) Fitzgerald, P. M. D. *J. Appl. Crystallogr.* **1988**, *21*, 273.
(11) Brünger, A. T. *X-PLOR Manual, Version* 2.1; Yale University, New Haven, 1990.

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