rangement at the terminus of migration thus is also highly stereospecific in the sense required by type-I geometry in the transition state.

The results form an instructive contrast with the stereochemistry of the regular dienyl 1,5-hydrogen shift. In the case of (*S*)-(2*E*,4*Z*)-6-methyl-2,4-octadiene-2-*d*,[18] for example, the electron distribution above and below the plane of the C_4–C_5 double bond (C_2–C_3 of the 1,5-system) of the diene is essentially isotropic, and suprafacial allowed reaction occurs on *both* faces of the receptor site at roughly equal rates. However, in the homodienyl case, where a cyclopropane unit replaces the C_2–C_3 double bond, the anisotropic electron distribution in the C_s-symmetric 3*E*′ ring orbital guides the flight of the hydrogen to only one stereochemical destination.

Acknowledgment. We thank the National Institute of General Medical Sciences for a grant in support of this work. E. J. Stark collaborated on the synthetic approach to lactol **9**.

Supplementary Material Available: Analytical, spectroscopic, and compositional charaterization of **5**, **12**, and the intermediates of Scheme I (18 pages). Ordering information is given on any current masthead page.

(18) Roth, W. R.; König, J.; Stein, K. *Chem. Ber.* **1970**, *103*, 426.

Persistence of Stereospecificity of Thermal Homodienyl Hydrogen Shift Reverse Ene Reactions in Cyclobutane Systems

Stephen J. Getty and Jerome A. Berson*

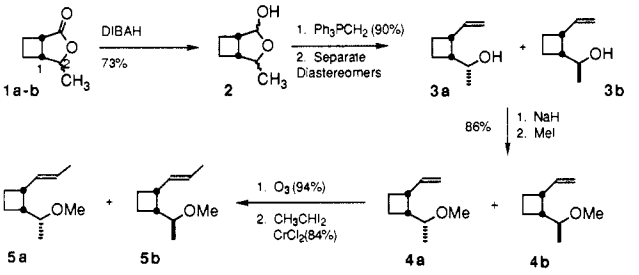
Department of Chemistry, Yale University
New Haven, Connecticutt 06511

Received September 15, 1989

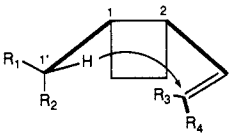
Stereospecificity survives in the thermal homo-Diels–Alder cycloreversions of 4,5-cyclobuta-3,4,5,6-tetrahydropyridazines despite the sharp diminution of the cycloreversion rate relative to the corresponding cyclopropanes.[1a,b] With the recent demonstration that a cyclopropane ring can control the stereochemistry of the 1,5-homodienyl hydrogen shift,[2] a comparison study is needed to test whether a cyclobutane can exert a similar influence in this sigmatropic rearrangement.[3]

In contrast to the *cis*-1-alkenyl-2-alkylcyclopropane case,[2] in which the only reaction was homodienyl hydrogen shift, the present *cis*-1-alkenyl-2-alkylcyclobutane experiment is complicated by the formation of several products, among which those resulting from homodienyl hydrogen shift constitute only about a fifth or less of the total. Moreover, the 1,5-diene structure of the product of the homodienyl hydrogen shift opens the possibility of further transformation by a Cope rearrangement. This problem can be suppressed by the choice of a methoxy group as a stereochemical marker, as in **4a,b** and **5a,b**, whose syntheses from the known[4] lactone **1** are described in Scheme I. Homodienyl hydrogen shift then leads to an enol ether whose subsequent Cope rearrangement is insignificant under these conditions.

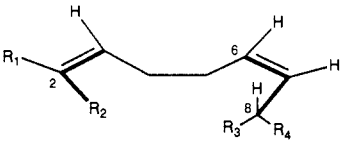
Scheme I



Upon being heated in the gas phase in basic lead glass ampoules[5] at 250 °C, compound **4b** disappeared at a first-order rate about



4a: R_1 = CH_3, R_2 = OMe, R_3, R_4 = H
4b: R_1 = OMe, R_2 = CH_3, R_3, R_4 = H
5a: R_1 = CH_3, R_2 = OMe, R_3 = D, R_4 = CH_3
5b: R_1 = OMe, R_2 = CH_3, R_3 = D, R_4 = CH_3



6: R_1 = CH_3, R_2 = OMe, R_3, R_4 = H
7: R_1 = OMe, R_2 = CH_3, R_3, R_4 = H
8: R_1 = OMe, R_2 = CH_3, R_3 = D, R_4 = CH_3
9: R_1 = CH_3, R_2 = OMe, C_8 config. not determined
10: R_1 = OMe, R_2 = CH_3, C_6 config. *E*, C_8 config. not determined

4×10^{-3} that of *cis*-2-isopropyl-1 *E*-propenylcyclopropane.[2] The respective Arrhenius parameters for **4a** and **4b** determined from measurements at 243.8, 255.9, and 267.5 °C were E_a = 47.8 ± 2 and 48.6 ± 2 kcal/mol and $A = 10^{14.8\pm0.9}$ and $10^{15.2\pm0.9}$ s^{-1}. The major products were formed by four pathways: (1) fragmentation to 1,3-butadiene and 2-methoxy-3-butene (35%); (2) epimerization at one or both of the ring stereogenic centers (35%); (3) [1,3]-sigmatropic rearrangement to 4-(1-methoxyethyl)cyclohexenes (10%); and (4) homodienyl hydrogen shift (retro-ene reaction) to 2-methoxy-2,6-octadienes (20%).

The retro-ene reactions strongly prefer the endo vinyl orientation, the sum of dienes **6** and **7** with the 6*Z*-configuration being 90% and 94%, respectively, of the total retro-ene product. Small amounts of 6*E*-dienes resulting from the stereochemical equivalent of reaction in the exo vinyl orientation are also formed, in contrast with the absence of such products in the cyclopropane series.[6] Within the endo manifold, the stereochemical course at the enol ether double bond also is highly stereospecific. The ratio **6**:**7** is about 26:1 from **4a**, and the inverse ratio **7**:**6** is about 56:1 from **4b**. These ratios are minimum estimates of the actual specificity of the hydrogen shifts in the endo manifold, since the competing double epimerization interconverts the two diastereomeric cyclobutane reactants. In fact, a computer-assisted simulation of the kinetic scheme shows that within experimental error, all of the minor 6*Z*-diene product in the endo manifold is accounted for by this side reaction. With respect to the rotational orientations of the receptor double bond and the alkyl hydrogen donor, the rearrangements of both diastereomers in the vinylcyclobutane series thus mimic the preference of the cyclopropane rearrangements[2] fo the overlap-favored geometry symbolized in **4a,b**/**5a,b**, despite the much slower rates in the cyclobutane systems. Since all of the stereospecificities in these and the previous[2] experiments are essentially off-scale, a quantitative comparison of the degree of stereochemical control by the cyclopropane and cyclobutane rings is not available.

The transition-state geometry deduced from these results

(1) (a) Berson, J. A.; Petrillo, E. W., Jr.; Bickart, P. *J. Am. Chem. Soc.* **1974**, *96*, 636. (b) Berson, J. A.; Olin, S. S.; Petrillo, E. W., Jr.; Bickart, P. *Tetrahedron* **1974**, *30*, 1639.

(2) Parziale, P. A.; Berson, J. A. *J. Am. Chem. Soc.*, preceding paper in this issue.

(3) A preliminary approach was made by Jordan and Berson (Jordan, L. M.; Berson, J. A. Unpublished work). (b) Jordan, L. M. Ph.D. Thesis, Yale University, New Haven, CT, 1973. (c) Reviewed by Gajewski, J. J. In *Hydrocarbon Thermal Isomerizations*; Academic Press: New York, 1981; p 178.

(4) Kosugi, H.; Sekiguchi, S.; Sekita, R.; Uda, H. *Bull. Chem. Soc. Jpn.* **1976**, *49*, 520.

(5) Doering, W. von E.; Beasley, G. H. *Tetrahedron* **1973**, *29*, 2231.

0002-7863/90/1512-1652$02.50/0

strongly implies the sense of stereogenicity transfer (1'*S* reactant → 8*S* product) from the origin to the terminus of hydrogen migration. This was confirmed independently (and with some difficulty) in the pyrolysis of the optically active isotopically labeled substrate 1*R*,2*R*,1'*S*-1-(1-methoxyethyl)-2-(2-*d*-1-*E*-propenyl)-cyclobutane (**5b**), which was obtained in 93% ee by the use of optically active (-)-(*S*)-3-butyn-2-ol for the synthesis of lactone **1b** (see Supplementary Material). At 239.4 °C, **5b** gave only about 5% of homodienyl shift products, of which the major component was the 2*E*, 6*Z* isomer (**8**, 90%). Minor components were the 2*Z*, 6*Z* (**9**, 4%) and the 2*E*, 6*E* (**10**, 6%). The 6*Z* products are those derived from an endo receptor orientation, and the observed 23:1 preference for the 2*E*, 6*Z* isomer in this manifold is consistent with an overlap-favored geometry. When hydrolyzed to the ketone and subjected to the same degradation procedure used in the cyclopropane work,[2] the mixed enol ethers **8-10** gave the *S*-2-*d*-propanoate of *R*-methyl mandelate in 81 ± 3% (^{2}H NMR analysis) and 69 ± 5% (^{1}H analysis, corrected for incomplete deuteration) of the diastereomeric excess maximally available from the ee level of the starting material. These again are minimum measures of the true stereospecificity.

At what ring size should stereospecificity of the homodienyl hydrogen shift disappear? If the hypothesis is correct that orbital overlap is the source of specificity here, even cyclopentane would be capable, in principle, of acting as a stereochemical control element, since one of its nearly degenerate HOMOs[7] has the appropriate[2] symmetry. For thermodynamic reasons, this would have to be studied experimentally in the forward ene rather than the reverse ene sense in most cases.

Acknowledgment. We thank the National Institute of General Medical Sciences for a grant in support of this work.

Supplementary Material Available: Description of and references for synthesis of isotopically labeled optically active reactant **5b** and characterization of substances of Scheme I and earlier synthetic intermediates (26 pages). Ordering information is given on any current masthead page.

(6) Reference 2 and references cited therein.
(7) Jorgensen, W. L.; Salem, L. *The Organic Chemist's Book of Orbitals*; Academic Press: 1973; p 256.

Novel Product from EPSP Synthase at Equilibrium

Gregory C. Leo, James A. Sikorski, and R. Douglas Sammons*

Monsanto Agricultural Company
A Unit of Monsanto Company, Technology Division
700 Chesterfield Village Parkway
Chesterfield, Missouri 63198

Received October 3, 1988
Revised Manuscript Received November 6, 1989

A novel shikimate ketal (**5**) is produced at equilibrium by the sixth enzyme in de novo aromatic amino acid biosynthesis, EPSPS[1] [EC 2.5.1.19].[2] The EPSP ketal is indisputably the species mistakenly labeled as an "enzyme free intermediate" by Barlow et al.[3] While **5** is produced at equilibrium, it clearly is not on the normal catalytic pathway. Understanding the formation of **5** is key to having a complete definition of the EPSPS chemical mechanism (Scheme I).

The EPSPS-bound intermediate (**4**) and **5** are readily apparent by ^{13}C NMR under internal equilibrium conditions.[3,12] When

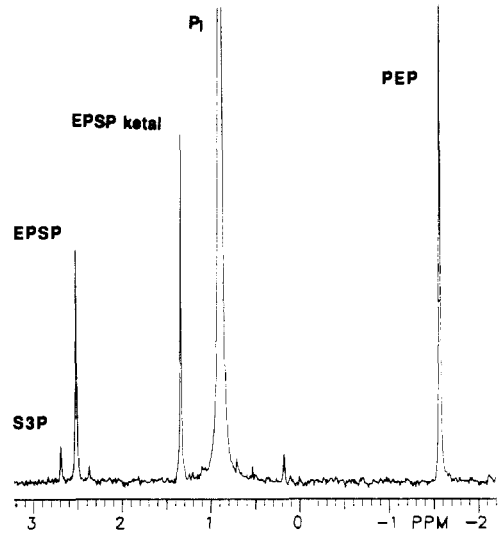
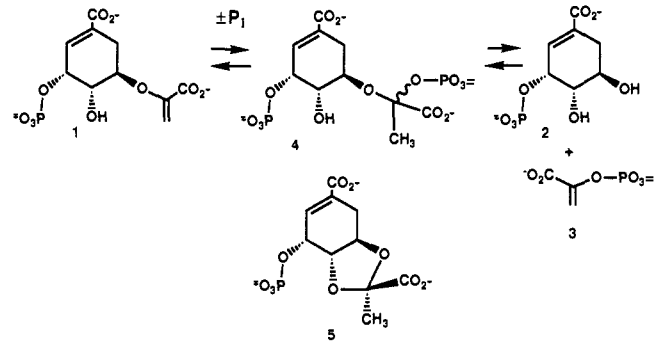


Figure 1. ^{31}P NMR spectrum of an 8-day internal equilibrium reaction with EPSPS set to maximize production of **5**, shown at 60% conversion. This mixture contained, after 8 days, 0.20 mM EPSPS, 0.1 M KP_i, pH 6.15, 14.7 mM **2**, 97.7 mM **3**, 9% glycerol, 47 mM KCl, 9 mM β-mercaptoethanol, 0.05 mM EDTA, 10 mg of Na_2WO_4, 10 mg of bacitracin, 10 mg of ampicillin, and 25 mg of trypsin inhibitor. The PEP was added intermittantly to drive the reaction as **1** was converted to **5** and **2** plus pyruvate. In 10 days, all of **2** was converted to **5**. Purification was carried out as described to remove excess PEP, pyruvate, and P_i to yield 1.1 g of the Na salt.[5] Spectra were taken with 0.75 mL of reaction mixture in 0.5 mL of D_2O on a Varian XL 300 instrument, 5-mm probe, ^{1}H decoupled, referenced to 1 N D_3PO_4.

Scheme I



(1) Abbreviations used: EPSPS, 5-enolpyruvoylshikimate 3-phosphate synthase; EPSP, 5-enolpyruvoylshikimate 3-phosphate; S3P, shikimate 3-phosphate; PEP, phosphoenolpyruvate; DTNB, 5,5'-dithiobis(2-nitrobenzoic acid); pD, proton activity in D_2O; [U-^{14}C], ^{14}C uniformly labeled; NOE, nuclear Overhauser effect; and P_i, inorganic phosphate.
(2) (a) Levin, J. G.; Sprinson, B. *J. Biol. Chem.* **1964**, *239*, 1142-1150. (b) Bondinell, W. E.; Vnek, J.; Knowles, P. E.; Sprecher, M.; Sprinson, B. *J. Biol. Chem.* **1971**, *246*, 6191-6196. (c) Amrhein, N.; Deus, B.; Gehrke, P.; Steinrücken, H. C. *Plant Physiol.* **1980**, *66*, 830-834.
(3) Barlow, P. N.; Appleyard, R. J.; Wilson, B. J. O.; Evans, J. *Biochemistry* **1989**, *28*, 7985-7991.
(4) Anderson, K. S.; Sikorski, J. A.; Johnson, K. A. *Biochemistry* **1988**, *27*, 7395-7406.
(5) Anion exchange on DEAE A-25 Sephadex with a linear gradient of TEAB from 0.4 to 1.0 M with **5** appearing at 0.75 M TEAB.
(6) CH_2N_2 (Arndt, F. *Org. Synth.* **1943**, *2*, 165-167) was added to a 0.5-mL MeOH/H_2O (4:1, v/v) solution of the salts precipitated with *p*-dioxane (5:1, v/v) from the HPLC eluent in footnote 5. The permethylated **1** and **5** (99% ^{13}C labeled at C'-2) were extracted with CH_2Cl_2 after removal of the organic layer with a nitrogen stream.
(7) ^{1}H NMR ($CDCl_3$): δ 6.74 (H-2, dd, *J* = 2.0, 5.1, 1 H), 5.03 (H-3, ddd, *J* = 4.6, 4.6, 7.3, 1 H), 4.125 (H-5, dd, *J* = 5.0, 10.0, 10.2, 1 H), 3.42 (H-4, dd, *J* = 4.2, 10.0, 1 H), 3.00 (H-6e, dd, *J* = 5.3, 16.3, 1 H), 2.385 (H-6a, ddd, *J* = 2.5, 10.6, 16.2, 1 H), 1.681 (CH_3, s, 3 H). ^{13}C NMR in D_2O (referenced to $CDCl_3$): δ 180.55 (q, *J* = 2.4), 178.34 (d, *J* = 5.8), 139.5 (C-1, ddd, *J* = 4.5, 7.4, 7.4), 134.2 (C-3, ddd, *J* = 4.4, 4.6, 164.5), 110.6 (C'-2, q, *J* = 4.5), 82.2 (dm, *J* = 146.6), 74.81 (dm, *J* = 155.4), 68.12 (d, *J* = 152.2), 34.42 (C-6, ddd, *J* = 8.1, 135.2, 135.2), 25.96 (C'-3, q, *J* = 128.5). ^{31}P NMR at pH 9.0 (1 N D_3PO_4/D_2O): δ 4.13 (d, *J* = 6.2).
(8) Castellano, S.; Leo, G.; Sammons, R. D.; Sikorski, J. A. *Biochemistry* **1989**, *28*, 3856-3868.
(9) Padgette, S. R.; Huynh, Q. K.; Akent, S.; Sammons, R. D.; Sikorski, J. A.; Kishore, G. M. *J. Biol. Chem.* **1988**, *263*, 1798-1802.

0002-7863/90/1512-1653$02.50/0