coupled activities of transketolase and DAHP synthase, the catalytic cornerstone of multistep immobilized enzyme synthesis of DAHP, increase carbon flow into aromatic amino acid biosynthesis, leading to increased synthesis of DAH and DAHP by microbial whole cells. Equally important, this synthesis is accomplished without the need for cofactor, cosubstrates, enzyme purification, enzyme immobilization, and adenosine triphosphate regeneration demanded by multistep enzymatic synthesis.

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(9) For examples where DAH has been used as a starting material in chemical syntheses, see: (a) Bartlett, P. A.; Satake, K. *J. Am. Chem. Soc.* **1988**, *110*, 1628. (b) Myrvold, S.; Reimer, L. M.; Pompliano, D. L.; Frost, J. W. *J. Am. Chem. Soc.* **1989**, *111*, 1861. See also ref 1a.

(10) Miller, J. H. *Experiments in Molecular Genetics*; Cold Spring Harbor Laboratory: Cold Spring Harbor, 1972.

Hemicarcerands Permit Entrance to and Egress from Their Inside Phases with High Structural Recognition and Activation Free Energies[1]

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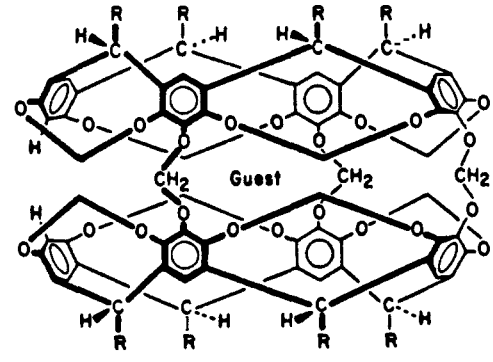
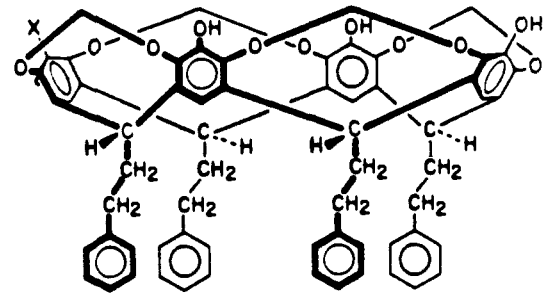
Previous papers reported that permanent guests were imprisoned during cavitand shell closures to form carceplexes,[2] which are closed-surface, hollow hosts that selectively incarcerate medium components (guests). This paper reports our first hemicarcerand (**1**), a carcerand[2] with a shell hole large enough to permit entrance and egress of molecule-sized guests (G), but which allows ordinary separations and characterizations of hosts and complexes.

Triol **2**[3] was isolated as byproduct (23%) in the synthesis of **3**.[2a] Shell closures of **2** were conducted identically with those for **3**[2a] (CH_2ClBr–K_2CO_3–solvent). Hemicarceplexes **1**·G were purified by chromatography on silica gel–$CHCl_3$/hexane and crystallized from $CHCl_3$–CH_3CN. Shell closures in $(CH_3)_2SO$ gave **1**·$(CH_3)_2SO$ (51%), in $(CH_3)_2NCOCH_3$ gave **1**·$(CH_3)_2NCOCH_3$ (42%), and in $(CH_3)_2NCHO$ gave **1**·$(CH_3)_2NCHO$ (20%). A stereoview of the crystal structure[4] of **1**·$(CH_3)_2NCHO$·$2CH_3CN$·$2CHCl_3$ is shown in **4**. Note that $(CH_3)_2NCHO$ is incarcerated. Each solvating CH_3CN is packed between each set of four $CH_2CH_2C_6H_5$ groups with N directed inward. Each $(CH_2CH_2C_6H_5)_4$–CH_3CN packet is capped with $CHCl_3$. The northern hemisphere in **4** is rotated about 20° with respect to the southern. The complex has a pseudo C_2 axis passing through the N and O atoms of $(CH_3)_2NCHO$, whose C=O group points toward the portal.

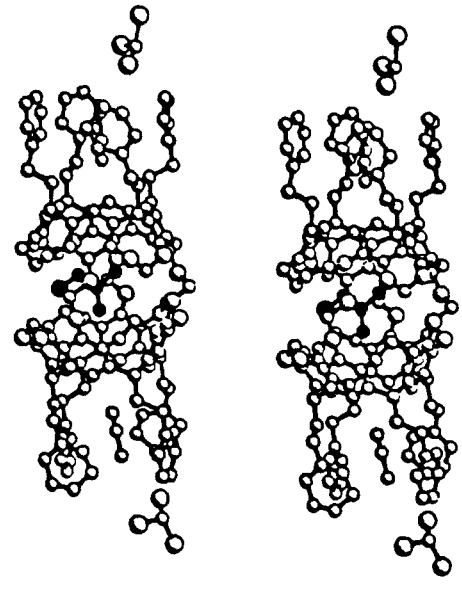
Chart I



1·G, R = $CH_2CH_2C_6H_5$



2, X = H; **3**, X = OH



4 (guest atoms of $(CH_3)_2NCHO$ darkened)

Heating hemicarceplexes in solvents too large to become guests gave **1**[5,6] by expelling guests: **1**·$(CH_3)_2SO$[5] required 214 °C for

(1) We warmly thank the National Science Foundation for supporting Grant NSF CHE 8802800.

(2) (a) Sherman, J. C.; Cram, D. J. *J. Am. Chem. Soc.* **1989**, *111*, 4527–4528. (b) Cram, D. J.; Karbach, S.; Kim, Y. H.; Baczyskyj, L.; Kalleymeyn, G. W. *Ibid.* **1985**, *107*, 2575–2576. (c) Cram, D. J.; Karbach, S.; Kim, Y. H.; Baczynskyj, L.; Marti, K.; Sampson, R. M.; Kalleymeyn, G. W. *Ibid.* **1988**, *110*, 2554–2560.

(3) New compounds gave elemental analyses within 0.40% of theory, the expected ^1H NMR, and FAB MS, M + 1 ions.

(4) Crystallization of **1**·$(CH_3)_2NCHO$ from $CHCl_3$–CH_3CN gave **1**·$(CH_3)_2NCHO$·$2CH_3CN$·$2CHCl_3$: orthorhombic, *Pbna* (standard setting *Pbcn*), a = 20.455 (5) Å, b = 20.773 (5) Å, c = 30.307 (8) Å, V = 12878 Å^3, Z = 4 (molecule has pseudo C_2 symmetry, the guest is disordered about a 2-fold axis, and the chloroform is disordered about an inversion center), R = 0.168. Details will be published elsewhere.

(5) Analyses for *all* elements present when summed came to 99.78–100.08%, individual analyses being within 0.40% of theory except for xenon in **1**·Xe (0.83% below 6.05% theory by thermal gravimetric analysis, summed analysis, 99.14%). Nitrogen analysis of **1** indicated that no nitrogen was present in the solid after drying at 70 °C for 12 h at 10^{-5} Torr.

(6) When a 5 mM solution of **1** in $CDCl_3$ was saturated at 25 °C with N_2 (5.6×10^{-3} M), ^1H NMR integrations of inward-turned intrahemisphere OCH_2O protons of the **1**·N_2 produced (δ, d, 3.97, 4.12 vs **1**, δ, d, 3.93, 4.09) gave a 1:1 ratio of species, which provides a K_a estimate of 180 M^{-1} for **1** + N_2 ⇌ **1**·N_2. In a similar experiment with O_2 (11.5 mM), a 1:2 ratio of **1**·O_2 to **1** was obtained, the inward OCH_2O protons of **1**·O_2 disappearing into the base line. The Ar_2CHR signals of **1** (δ, t, 4.80 and t, 4.90) were broadened and moved to δ, 5.24 and 5.38 in **1**·O_2, and their integrals were used in the K_a estimate of 44 M^{-1} for **1** + O_2 ⇌ **1**·O_2.

0002-7863/90/1512-1659$02.50/0

48 h in 1,2,4-$Cl_3C_6H_3$; 1·$(CH_3)_2NCOCH_3$,[5] 165 °C for 24 h in 1,3,5-$(CH_3)_3C_6H_3$; 1·$(CH_3)_2NCHO$,[5] 165 °C for 12 h. In the 360-MHz ^1H NMR spectra, proton signals of incarcerated guests are far upfield of guests simply dissolved in $CDCl_3$: 1·$(CH_3)_2SO$, δ -1.02; 1·$(CH_3)_2NCOCH_3$, δ -2.30, -1.33, 1.05; 1·$(CH_3)_2NCHO$, δ -1.04, -0.21, and 4.14. Proton signals of the northern and southern hemispheres of the host are identical at 25 °C for **1**, for 1·$(CH_3)_2SO$, and for 1·$(CH_3)_2NCHO$, but different for 1·$(CH_3)_2NCOCH_3$. Thus end-to-end guest rotation relative to the host's north-south axis is inhibited only in 1·$(CH_3)_2NCOCH_3$.

Treating free **1** in appropriate solvents gave new hemicarceplexes, e.g.: 1·CH_3CN[3] (δ -2.42); 1·CS_2,[3] 1·pyridine[5] (heat required); 1·CH_2Br_2.[3] When treated with a 0.14 M solution of xenon in $CDCl_3$ at 25 °C, 1·Xe[5] formed, whose 1·^{129}Xe NMR signal was at -101 ppm (dissolved xenon, 0 ppm).[7] On silica gel -15% hexane/85% $CHCl_3$ (v/v), most complexes (and free **1**) had different R_f (TLC) values. These one-to-one complexes (^1H NMR proton counting and elemental analyses) were stable to laboratory manipulations at room temperature, but released their guests when subjected to FAB MS to give strong M + 1 signals for **1**.[7,8]

These results demonstrate that hemicarcerands can be designed and prepared whose portals show high structural recognition in guest entry, departure, and residence. We envision potential uses for hemicarceplexes: drug delivery systems; organ imaging; protection of bone from deposition of heavy metal salts useful in radiation therapy; light switches; information storage. We are examining these possibilities in many carceplexes.

(7) A k_{in} second-order rate constant for filling **1** (5 mM) with Xe (0.14 M) in $CDCl_3$ at 25 °C was estimated to be 0.055 min^{-1} M^{-1} (followed by ^1H NMR changes). A k_{out} first-order rate constant estimate in CD_2Cl_2 at 25 °C for 1·Xe + CD_2Cl_2 → 1·CD_2Cl_2 + Xe (followed by ^1H NMR changes) gave 2.5×10^{-4} min^{-1}. If we assume that k_{out} in CD_2Cl_2 ~ k_{out} in $CDCl_3$, K_e for 1 + Xe ⇄ 1·Xe is estimated to be ≈200 M^{-1}.

(8) The closest precedents to our hemicarcerands are the elegant *cryptophanes* of A. Collet (summarized in *Tetrahedron* **1987**, *43*, 5725-5759), who joined two cyclotriveratrylene-like modules with three ethylene bridges to give hollow molecules with three equivalent portals.

On the Mechanism of Action of Vitamin K. A New Nonenzymic Model

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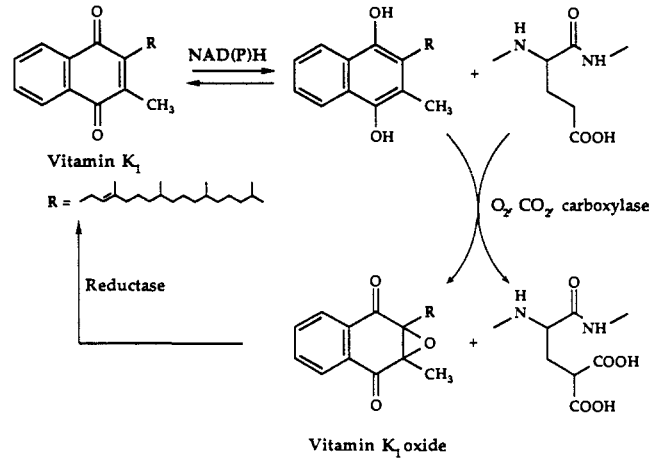
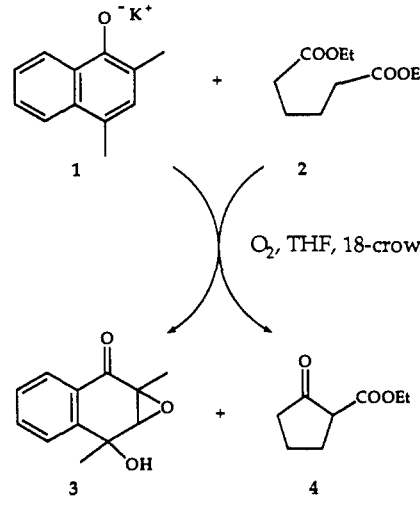
Vitamin K is essential for blood clotting.[1] It may also play a role in bone calcification[2] and have other broad functions in biological systems.[3] At the enzyme level, vitamin K is an obligatory cofactor promoting the posttranslational carboxylation[4] of selected glutamic acid residues in many of the proteins of the blood clotting cascade, including factor II (prothrombin), factor VII, factor IX, factor X, protein C, protein M, protein S, and protein Z, as well as the bone protein osteocalcin.[5]

The carboxylative conversion of glutamate to γ-carboxyglutamate requires the hydroquinone form of vitamin K (or vitamin K and NAD(P)H), oxygen, carbon dioxide, and a membrane-bound carboxylase[1] only recently isolated in pure form.[6] In the course of the carboxylation (Scheme I), vitamin K is converted to vitamin K oxide.[7] A second, reductase-catalyzed pathway returns vitamin K oxide to vitamin K for a new catalytic cycle.[8,9]

Scheme I



Scheme II



In coming to grips with the mechanism of action of vitamin K, it is important to establish whether the formation of vitamin K oxide is an integral part of the step that effects the carboxylation.[10] Through the recent efforts of Suttie and his collaborators,[11] it has been shown that the degree of carboxylation closely parallels the extent of formation of vitamin K oxide under diverse circumstances.[6]

Since the discovery of the vitamin K dependent carboxylation, a variety of mechanistic proposals have been advanced ranging from free-radical[12,13] to base-promoted pathways.[14-17] Missing

(1) Recent reviews: (a) Suttie, J. W. *Biofactors* **1988**, *1*, 55. (b) Suttie, J. W. *Annu. Rev. Biochem.* **1985**, *54*, 459. (c) Olson, R. E. *Annu. Rev. Nutr.* **1984**, *4*, 281.

(2) Price, P. A. *Annu. Rev. Nutr.* **1988**, *8*, 565.

(3) Biggins, J.; Mathis, P. *Biochemistry* **1988**, *27*, 1494.

(4) Stenflo, J. *J. Biol. Chem.* **1974**, *249*, 5527. Nelsestuen, G. L.; Zytkovicz, T. H.; Howard, J. B. *J. Biol. Chem.* **1974**, *249*, 6347. Magnusson, S.; Sottrup-Jensen, L.; Petersen, T. E.; Morris, H. R.; Dell, A. *FEBS Lett.* **1974**, *44*, 189.

(5) Hauschka, P. V.; Lian, J. B.; Gallop, P. M. *Proc. Natl. Acad. Sci. U.S.A.* **1975**, *72*, 3925. Price, P. A.; Otsuka, A. S.; Poser, J. W.; Kirstaponis, J.; Raman, N. *Proc. Natl. Acad. Sci. U.S.A.* **1975**, *73*, 1447.

(6) Hubbard, B. R.; Ulrich, M. M. W.; Jacobs, M.; Vermeer, C.; Walsh, C.; Furie, B.; Furie, B. C. *Proc. Natl. Acad. Sci. U.S.A.* **1989**, *86*, 6893.

(7) Matschiner, J. T.; Bell, R. G.; Amelotti, J. M.; Knauer, T. E. *Biochim. Biophys. Acta* **1970**, *201*, 309.

(8) Bell, R. G.; Matschiner, J. T. *Arch. Biochem. Biophys.* **1970**, *141*, 473. Fieser, L. F.; Tishler, M.; Sampson, W. L. *J. Biol. Chem.* **1941**, *137*, 659. Sadowski, J. A.; Suttie, J. W. *Biochemistry* **1974**, *13*, 3696. Preusch, P. C.; Suttie, J. W. *J. Biol. Chem.* **1983**, *258*, 714.

(9) Silverman, R. B. *J. Am. Chem. Soc.* **1981**, *103*, 5939. Preusch, P. C.; Suttie, J. W. *J. Org. Chem.* **1983**, *48*, 3301.

(10) Willingham, A. K.; Matschiner, J. T. *Biochem. J.* **1974**, *140*, 435.

(11) Sadowski, J. A.; Schnoes, H. K.; Suttie, J. W. *Biochemistry* **1977**, *16*, 3856. Larson, A. E.; Friedman, P. A.; Suttie, J. W. *J. Biol. Chem.* **1981**, *256*, 11032. Wallin, R.; Suttie, J. W. *Arch. Biochem. Biophys.* **1982**, *214*, 155. Wood, G. M.; Suttie, J. W. *J. Biol. Chem.* **1988**, *263*, 3234.

(12) Gallop, P. M.; Friedman, P. A.; Henson, E. M. *Vitamin K Metabolism and Vitamin K Dependent Proteins*; Suttie, J. W., Ed.; University Park Press: Baltimore, MD, 1980; pp 408-412.

(13) For a working example of a free-radical carboxylation of glycine, see: Wheelan, P.; Kirsch, W. M.; Koch, T. H. *J. Org. Chem.* **1989**, *54*, 4360.

(14) DeMetz, M.; Soute, B. A.; Hemker, H. C.; Fokkens, R.; Lugtenberg, J.; Vermeer, C. *J. Biol. Chem.* **1982**, *257*, 5326.

(15) Suttie et al. have proposed a 2-hydroperoxy vitamin K intermediate: Suttie, J. W.; Larson, A. E.; Canfield, L. M.; Carlisle, T. L. *Fed. Proc. Am. Soc. Exp. Biol.* **1978**, *37*, 2605.

0002-7863/90/1512-1660$02.50/0