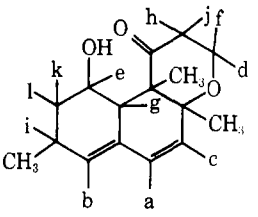

and chemical shifts were recorded relative to TMS. On the basis of chemical shift, multiplicity, and proximity to electronegative functionality, the assignments shown in Table I were made.[4-6]

Table I. ^{13}C Chemical Shifts of I in $CDCl_3$ Relative to TMS Together with Multiplicities and Assignments

δ_C	Multiplicity	Carbon no. assignment
195.9	s	11
135.5	d	4, 6, 7
132.1	d	
128.2	d	
130.0	s	5
78.7	s	8
66.8	d	1
60.0	t	13
57.0	s	9
41.7	d	10
39.3	t	12
38.6	t	2
25.3	d	3
21.1	q	methyls at 3, 8, and 9
20.7	q	
13.0	q	

The chirality of simple, optically active, transoid, heteroannular, conjugated dienes correlates well with the Cotton effects of their $\pi \rightarrow \pi^*$ transitions in the 230–280-nm region.[7]

The work of Beecham, *et al.*,[8,9] has shown that the presence of an allylic oxygen as in I dramatically affects the applicability of the basic diene helicity rule. According to Beecham, the "oxygen forming helix" or the C=CCO system determines the sign of the observed Cotton effect.[10] The CD curve of I is a double humped curve with $\Delta\epsilon_{242.5} = -21.9$ and $\Delta\epsilon_{212.5} = +21.9$; consequently, the stereochemistry at C8 is defined with the C=CCO system having left-handed helicity and the methyl group axial.

When I was subjected to the Horeau procedure,[11] the recovery of (−)-α-phenylbutyric acid (R-acid) determines the configuration at C1 to be S. In addition, since the hydroxyl and carbonyl groups are chelated and since protons H_e and H_g are cis to one another (J_{eg} = 4.0 Hz) the stereochemistry at C1 and C9 is defined so that the methyl group is axial and the pyranone ring is in the chair conformation.

(4) E. Wenkert and B. L. Buckwalter, *J. Amer. Chem. Soc.*, **94**, 4367 (1972).
(5) J. Polonsky, Z. Baskevitch, N. Cagnoli-Bellavita, P. Ceccherelli, B. L. Buckwalter, and E. Wenkert, *J. Amer. Chem. Soc.*, **94**, 4369 (1972).
(6) P. S. Steyn, P. L. Wessels, C. W. Holzapfel, D. J. J. Potgieter, and W. K. A. Louw, *Tetrahedron*, **28**, 4775 (1972).
(7) E. Charney, H. Ziffer, and U. Weiss, *Tetrahedron*, **21**, 3121 (1965).
(8) A. F. Beecham, *Tetrahedron*, **27**, 5207 (1971).
(9) A. F. Beecham, A. McL. Mathieson, S. R. Johns, J. A. Lamberton, A. A. Sioumis, T. J. Batterham, and I. G. Young, *Tetrahedron*, **27**, 3725 (1971).
(10) Dreiding models of I indicate that with the half-chair conformations of the decalin rings the diene system is virtually planar.
(11) A. Horeau and H. B. Kagan, *Tetrahedron*, **20**, 2431 (1964).

In the pmr spectrum of I, taken in pyridine, the broad signal of H_i is located at δ 2.65 which is a deshielding shift of δ 0.15 from that observed in $CDCl_3$. This means that this proton is 1,3-diaxial relative to the hydroxyl group[12] and the secondary methyl is thus equatorial as expected. Hence the stereochemistry of I is completely defined.

The unusual constitution of I may be formally derived from a polyketide intermediate which consists of five acetate and two propionate units.[13]

The presence of a tertiary methyl group such as that at C9 is rare in polyketide derived mold metabolites. To our knowledge, the only example is portentol,[2] although it seems reasonable to suggest that diplodiatoxin[6] is another example.

(12) P. V. Demarco, E. Farkas, D. Doddrell, B. L. Mylari, and E. Wenkert, *J. Amer. Chem. Soc.*, **90**, 5480 (1968).
(13) It is entirely possible that the C3 and C9 methyls are introduced from the one carbon pool. This would preserve the notion that no fungal product incorporates propionate within the chain as pointed out by Turner (see ref 2, p 363).

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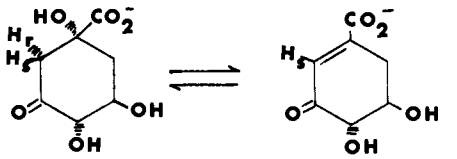
Mechanism of Dehydroquinase Catalyzed Dehydration. I. Formation of a Schiff Base Intermediate

Sir:

The conversion of dehydroquinic acid (**1**) to dehydroshikimic acid (**2**), is catalyzed by the enzyme dehydro-



1 **2**

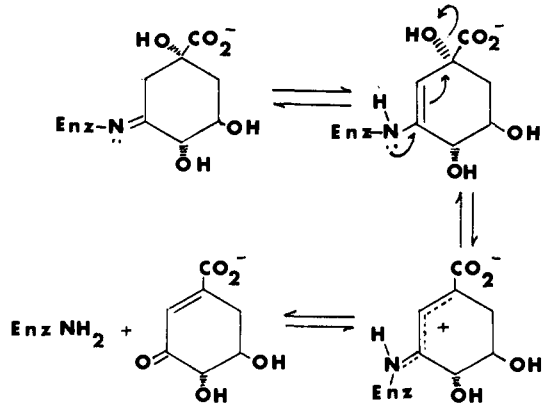
lyase (5-dehydroquinate hydrolyase, E C 4.2.1.10). Dehydroquinase was first isolated and partially purified by Mitsuhashi and Davis,[1] and was subsequently used in the classic determination of the absolute stereochemical course of citric acid biosynthesis by Hanson and Rose.[2] These researchers also established that the elimination of water from **1** proceeds in a syn manner so that the prochiral *R* proton is eliminated under equilibration conditions in contrast to the anti elimination most frequently observed in other carbon–oxygen lyase systems.[3] Rose has suggested that these rarely observed enzymatic syn eliminations can be reasonably explained in terms of carbanion intermediates.[2,3] We present evidence here that this syn dehydration involves Schiff base formation between the enzyme and its substrate **1**.

Dehydroquinase was isolated from *E. coli* 83-2[4] and purified according to published procedures.[1] Succes-

(1) S. Mitsuhashi and B. D. Davis, *Biochim. Biophys. Acta*, **15**, 54 (1954).
(2) K. R. Hanson and I. A. Rose, *Proc. Nat. Acad. Sci. U. S.*, **50**, 981 (1963).
(3) I. A. Rose in "The Enzymes," Vol. II, 3rd ed, P. D. Boyer, Ed., Academic Press, New York, N. Y., 1970, p 309.
(4) An initial strain of these bacteria was kindly furnished by Professor B. D. Davis and P. C. Tai.

sive ammonium sulfate precipitations and calcium phosphate gel treatment produced an enzyme preparation in which the specific activity was 20-fold greater than previously reported.[1] To a cold (0°) solution of dehydroquinase (1.92 units/mg) in 5.0 ml of Tris buffer (pH 7.4, 0.03 *M*) was added 1.0 ml of 0.76 *M* sodium borohydride in water over a period of 30 min. The pH was maintained between 7.5 and 8.0 by the dropwise addition of 0.5 *N* acetic acid. After dialysis against Tris buffer, the activity of the enzyme was 1.65 units/mg. Another solution containing substrate **1**[5] (0.735 mmol) and enzyme was treated with sodium borohydride in an identical manner. After dialysis the specific activity of the enzyme isolated from this solution was essentially inactive with a maximum activity of 0.0272 unit/mg.[6]

Deactivation of enzymes by sodium borohydride only in the presence of substrate is diagnostic of a Schiff base intermediate in enzymatic systems.[7-9] We therefore propose that the dehydration occurs *via* Schiff base formation between the enzyme and **1** as formulated below.



Acknowledgments. We thank Professors W. C. McDonald and J. R. Seed and Messrs. M. Dureau and M. Fontane for their help in growth and maintenance of bacteria. This research was supported in part by a National Institute of Health Biomedical Sciences Support Grant administered by Tulane University.

(5) E. Haslam, R. D. Haworth, and P. F. Knowles, *Methods Enzymol.*, 6, 498 (1963).

(6) Control experiments have shown that enzymatic activity is not lost when the enzyme is incubated with 0.735 mmol of substrate **1** in the absence of sodium borohydride.

(7) E. Grazi, T. Cheng, and B. L. Horecker, *Biochem. Biophys. Res. Commun.*, 7, 250 (1962).

(8) B. L. Horecker, S. Pontremoli, C. Ricci, and T. Cheng, *Proc. Nat. Acad. Sci. U. S.*, 47, 1949 (1961).

(9) Reference 3, p 335 ff.

(10) National Institutes of Health Career Development Award Recipient 1972-1977.

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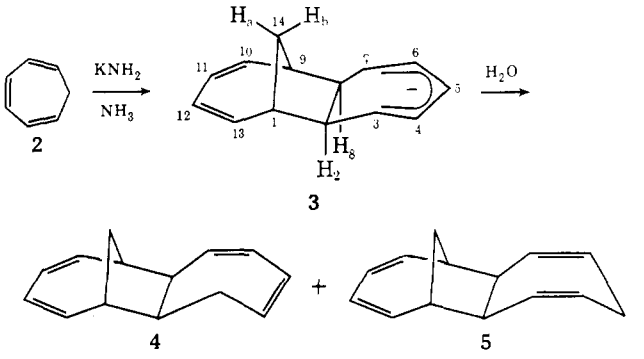
Mechanism of the Base-Promoted Cyclodimerization of Cycloheptatriene

Sir:

Theoretical considerations suggest that the cycloheptatrienyl anion (**1**), one of the simplest "antiaromatic" carbocyclic π systems, will suffer Jahn–Teller distortion from a triplet state of D_{7h} symmetry to one or more lower symmetry and lower energy singlet states.[1] Because of the possible availability of a variety of thermally accessible states, the chemical behavior of this highly reactive species represents an interesting problem.[2,3] We now report a remarkably rapid and highly regio- and stereoselective base-promoted cyclodimerization of cycloheptatriene and present evidence for the intermediacy of the cycloheptatrienyl anion (**1**) in this novel process.

When cycloheptatriene (**2**) is added to a 0.7 *M* solution of potassium amide in liquid ammonia at −33° a deep red-brown color is formed immediately. Upon quenching into saturated aqueous ammonium chloride–pentane after 10 min a mixture of cycloheptatriene dimers consisting predominantly of **4** (81%) and **5** (16%) was obtained. Isolated yields were as high as



88%. The nmr spectrum of the initial red-brown solution, obtained at −55° in ammonia-d_3,[4] is completely consistent with anion **3**. The spectral parameters for **3** (Table I) are closely analogous to those for

Table I. Nmr Spectral Data for Tricyclo[7.4.1.0^{2,8}]tetradeca-3,5,10,12-tetraen-7-ylpotassium (**3**) in Ammonia-d_3 at −55°

Proton(s)[a]	Chemical shift (ppm)[b]	Proton(s)[a]	Chemical shift (ppm)[b]
H_1, H_9	2.25	H_{10}, H_{13}	~6.2
H_2, H_8	3.17	H_{11}, H_{12}	~5.8
H_3, H_7	3.75	H_{14a}	0.80
H_4, H_6	5.77	H_{14b}	3.03
H_5	3.18		

[a] $J_{1,13} \approx J_{1,14b} \approx 6.9$; $J_{23} = J_{78} = 4.0$; $J_{34} = J_{67} = 10.5$; $J_{45} = J_{56} = 8.0$; $J_{35} = J_{57} = 1.2$; $J_{12} \approx J_{89} \approx 0$ Hz; $J_{14a,14b} \approx 12$ Hz. [b] Trimethylamine ($\delta^{NH_3}_{TMS} = 2.135$) was used as an internal standard.

bicyclo[4.2.1]nona-2,4-diene,[5] bicyclo[4.2.1]nona-2,4,7-

(1) (a) R. Breslow and H. W. Chang, *J. Amer. Chem. Soc.*, **84**, 1484 (1962); **87**, 2200 (1965); (b) L. C. Snyder, *J. Phys. Chem.*, **66**, 2299 (1962); (c) M. J. S. Dewar and N. Trinajstić, *Tetrahedron*, **26**, 4269 (1970).

(2) For studies of anion **1**, see (a) H. J, Dauben, Jr., and M. R. Rifi, *J. Amer. Chem. Soc.*, **85**, 3041 (1963); (b) W. von E. Doering and P. P. Gaspar, *ibid.*, **85**, 3043 (1963); (c) R. Breslow and W. Chu, *ibid.*, **95**, 411 (1973).

(3) For studies of derivatives of **1**, see ref 1a and (a) S. W. Staley and A. W. Orvedal, *J. Amer. Chem. Soc.*, **95**, 3382 (1973); (b) K. Takahashi, H. Yamamoto, and T. Nozoe, *Bull. Chem. Soc. Jap.*, **43**, 200 (1970).

(4) I. Melczynski, *Angew. Chem.*, **74**, 32 (1962). We thank Dr. John Morrison for assistance with the preparation of ammonia-d_3.

(5) C. W. Jefford and F. Delay, *J. Amer. Chem. Soc.*, **94**, 4794 1972) C. W. Jefford, U. Barger, and F. Delay, *Helv. Chim. Acta*, **56**, 1083 (1973). We thank Professor Jefford and Dr. Delay for a copy of the nmr spectrum of this compound.