be visualized. Whatever its structure, the intermediate possesses the interesting ability to generate products with, and without, bond formation between the β carbon atoms of the original cross-conjugated dienone system of **1**. Further characterization of this species is the goal of our continuing research.[10]

Acknowledgment. This work was supported by the U. S. Public Health Service.

(10) Hydroxylic solvent adducts similar to those described herein have been recently reported for 2,6-cycloheptadienone: H. Nozaki, M. Kurita, and R. Noyori, *Tetrahedron Lett.*, 3635 (1968).

(11) (a) Alfred P. Sloan Research Fellow, 1968–1970; (b) NASA Predoctoral Fellow, 1965–1968.

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Contribution No. 1612, Department of Chemistry
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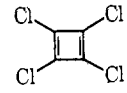
Received August 2, 1968

Interception of Transient Tetrachlorocyclobutadiene[1]

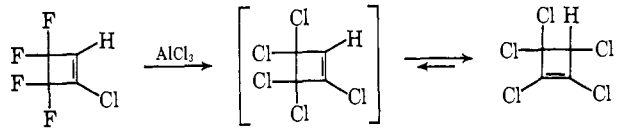
Sir:

We report here the generation and trapping of a reactive species which we believe is tetrachlorocyclobutadiene (I). Reaction of 2-chloro-3,3,4,4-tetra-



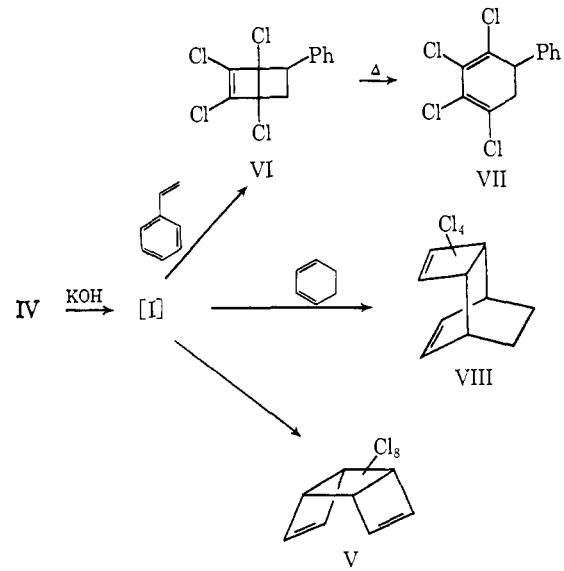
I

fluorocyclobutene[2] (II) with aluminum trichloride in dichloromethane[3] gave, as the sole product in 74–86% yield, 3-H-pentachlorocyclobutene[4] (IV), $\nu_{C=C}$ 1637 cm^{-1}, τ 4.78 (s). None of the 1-H isomer (III) was detected in the reaction mixture although the conditions would undoubtedly cause equilibration of III and IV; the greater stability of IV was expected by analogy[5] but stands in contrast to the behavior of trichlorocyclopropene, where the 1-H isomer is favored.[6] Compound III rearranges to 1-H-pentachlorobutadiene-1,3[7] above *ca.* 160°, but may conveniently be purified by vacuum distillation through a spinning-band[8] column, bp 89–90° (19 mm).



II III IV

Reaction of compound IV with an excess of powdered 85% potassium hydroxide in refluxing benzene, best with high-speed stirring, effects dehydrohalogenation and in the absence of a reactive diene or dienophile gives up to 43% of a compound C_8Cl_8,[4] assigned structure V. If excess styrene is present, I is trapped as its Diels–Alder adduct VI,[4] formed in 18–20% yield along with a comparable amount of V. In the presence of excess cyclohexa-1,3-diene, adduct VIII[4] is formed in 22% yield, accompanied by traces of V. The structure



VI VII VIII V

of VI, mp 35.5–37°, $\nu_{C=C}$ 1603 cm^{-1}, follows from its composition, its ir and nmr spectra, and its ready thermal isomerization to the known 1,2-dihydro-3,4,5,6-tetrachlorobiphenyl[9] (VII) identified by comparison (ir spectra, mixture melting point) with an authentic sample. Only a single isomer of VI has been found in the reaction mixture, but we have no evidence for the stereochemistry of the phenyl group.

The structure of VIII, mp 101–103°, $\nu_{C=C}$ 1626 cm^{-1}, was assigned largely on the basis of its nmr spectrum. Three centrosymmetric multiplets are observed at 3.84 (2 H), 7.15 (2 H), and 7.6–8.8 (4 H), and the vinyl proton signal at lowest field collapses to a singlet on irradiation of the τ 7.15 peak. The *syn* disposition of the four-membered ring is assigned by analogy (Alder rule).

The structure of V, mp 161–163° dec, was initially assigned from its spectroscopic properties: ir, $\nu_{C=C}^{C_2Cl_4}$ 1606 and 1580 cm^{-1}, with other medium-to-strong bands at 1164, 1060, 1012, 991, 792, 663, and 596 cm^{-1} (in CS_2); uv, λ_{max}^{EtOH} 238 mμ (ϵ 5700). The ultraviolet spectrum is inconsistent with either non-interacting double bonds (in the *anti* isomer)—*cf.* λ_{max} 210 mμ (ϵ 1000) for IV—or a perchlorinated conjugated diene—*cf.* λ_{max} 287 mμ (ϵ 7000) for VII—and may be rationalized by postulating excited-state interaction of the adjacent but nonconjugated double bonds. An X-ray study of V has been undertaken by Dodge and Templeton, who report[10] good (R = 9%) refinement to octachlorocubane! Examination of a molecular model of V indicates that the eight chlorines lie very near the corners of a cube, and we believe the X-ray results are consistent with packing of V in the cubane lattice with random orientation of the C_2 axis

(1) Presented in part at the 155th National Meeting of the American Chemical Society, San Francisco, Calif., April 3, 1968, Abstract P180. Grateful acknowledgment is made to the donors of the Petroleum Research Fund and the National Institutes of Health for grants (PRF 2191-A1,4, GM-12731, and GM-15678) in partial support of this work.

(2) D. J. Burton and R. L. Johnson, *J. Amer. Chem. Soc.*, **86**, 5361 (1964).

(3) W. C. Soloman, L. A. Dee, and D. W. Schultz, *J. Org. Chem.*, **31**, 1551 (1966).

(4) Elemental composition of new compounds was established by satisfactory C, H, and Cl analyses and by mass spectrometry.

(5) J. D. Park, J. R. Lacher, and J. R. Dick, *J. Org. Chem.*, **31**, 1116 (1966).

(6) R. Breslow and G. Ryan, *J. Amer. Chem. Soc.*, **89**, 3073 (1967); R. Breslow, paper presented at the International Symposium on the Chemistry and Applications of Small Ring Compounds, Louvain, Belgium, Sept 1967.

(7) A. Roedig, R. Kohlhaupt, and G. Markl, *Ber.*, **99**, 698 (1966).

(8) Auto-Annular still, Nester–Faust Manufacturing Corp. Newark, Del.

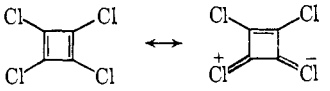
(9) E. T. McBee, W. R. Diveley, and J. E. Burch, *J. Amer. Chem. Soc.*, 77, 385 (1955).

(10) R. P. Dodge and D. H. Templeton, private communication.

among the six possible directions and consequent observation of averaged atomic coordinates. The possibility of the molecule having different structures in the solid state and in solution is ruled out by nearly identical solution and KBr pellet ir spectra.

The stereospecificity of the additions observed so far and the effectiveness of electron-rich[11] olefins in intercepting the intermediate lead us to postulate that the reactive species is singlet tetrachlorocyclobutadiene. A control experiment showed that cyclohexadiene does not add to IV under the reaction conditions, and we would not expect the pentachlorocyclobutenyl anion to add to either styrene or cyclohexadiene.

This strikingly straightforward synthesis of a substituted cyclobutadiene probably rests heavily on the ability of chlorine both to accept and to donate electron density, with resulting "push–pull"[12] stabilization as indicated.



Further work on the preparation and transformations of V and its adducts is in progress.

(11) J. Sauer and H. Wiest, *Angew. Chem. Intern. Ed. Engl.*, **1**, 269 (1962).

(12) R. Breslow, D. Kivelevich, M. J. Mitchell, W. Fabian, and K. Wendel, *J. Amer. Chem. Soc.*, **87**, 5132 (1965).

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Separation of the Cyclization and Rearrangement Processes of Sterol Biosynthesis. Enzymic Formation of a Protosterol Derivative

Sir:

The biosynthesis of sterols is presumed to involve cyclization of 2,3-oxidosqualene[1,2] to the cation **1** (or its functional equivalent) and subsequent rearrangement by a sequence of 1,2 shifts to lanosterol (**2**).[3,4] The driving force for the over-all rearrangement comes at least in part from the relief of repulsive interactions in **1** which are associated with the B ring (obligatory twist-boat geometry) and its substituents, especially the methyl group attached to C-8. This communication reports the results of a study of the action of 2,3-oxidosqualene–sterol cyclase on the unnatural substrate **3** which by the normal mode of cyclization would produce the cation **4**, a structure lacking the methyl substituents at C-8 and C-14 of **1** and, therefore, less strained and less apt to rearrange to a lanosterol analog. This study demonstrates that the structure (**4**) produced by cyclization of the substrate **3** undergoes proton elimination to form the *unrearranged protosterol derivative* **10**.[5]

(1) (a) E. J. Corey and W. E. Russey, *J. Amer. Chem. Soc.*, **88**, 4751 (1966); (b) E. J. Corey, W. E. Russey, and P. R. Ortiz de Montellano, *ibid.*, **88**, 4750 (1966); (c) W. E. Russey, Ph.D. Thesis, Harvard University, 1966.

(2) (a) E. E. van Tamelen, J. D. Willet, R. B. Clayton, and K. E. Lord, *J. Amer. Chem. Soc.*, **88**, 4752 (1966); (b) J. D. Willet, K. B. Sharpless, K. E. Lord, E. E. van Tamelen, and R. B. Clayton, *J. Biol. Chem.*, **242**, 4182 (1967).

(3) R. B. Woodward and K. E. Bloch, *J. Amer. Chem. Soc.*, **75**, 2023 (1953).

(4) For a comprehensive review see R. B. Clayton, *Quart. Rev.* (London), **19**, 168 (1965).

(5) The term protosterol is used here to mean the tetracyclic system corresponding to the presently hypothetical precursor **1**. Fusidic acid [W. O. Godtfredsen, W. von Daehne, S. Vangedal, A. Marquet, D. Arigoni, and A. Malera, *Tetrahedron*, **21**, 3505 (1965)], helvolic acid [S. Okuda, S. Iwasaki, M. I. Sair, Y. Machida, A. Inoue, and K. Tsuda, *Tetrahedron Lett.*, 2295 (1967)], and cephalsporin P_1 [T. G. Halsall, E. R. H. Jones, G. Lowe, and C. E. Newall, *Chem. Commum.*, 685 (1966)] represent specific natural products in the protosterol (fusidane) series.

(6) E. E. van Tamelen and T. J. Curphey, *Tetrahedron Lett.*, 121 (1962).

(7) E. J. Corey, J. A. Katznellenbogen, and G. H. Posner, *J. Amer. Chem. Soc.*, **89**, 4245 (1967).

(8) J. M. Osbond, *J. Chem. Soc.*, 5270 (1961).

(9) See E. J. Corey, M. F. Semmelhack, and L. S. Hegedus, *J. Amer. Chem. Soc.*, **90**, 2416 (1968), and references therein cited.

(10) The coupling process produced a mixture of three geometrical isomers, differing at the two central (disubstituted) olefinic linkages, *trans,trans* (desired product, **5**), *trans,cis*, and *cis,cis* in a ratio of 90:9:1 [gas chromatographic (gc) analysis using Epon 1001 on Diatoport S (F & M Co.) as stationary phase].

(11) Infrared, nuclear magnetic resonance, and mass spectra of **3** and **5** were fully consistent with the assigned structures.

(12) E. J. Corey, K. Lin, and M. Jautelat, *J. Amer. Chem. Soc.*, **90**, 2724 (1968).

(13) The solution of enzyme was prepared by a modification of the method previously described [P. D. G. Dean, P. R. Ortiz de Montellano, K. Bloch, and E. J. Corey, *J. Biol. Chem.*, **242**, 3014 (1967)] which has been developed in these laboratories by Dr. Shozo Yamamoto; see P. Ortiz de Montellano, Ph.D. Thesis, Harvard University, 1968, p 106.

The unlabeled oxido derivative **3** was synthesized by terminal epoxidation[6] of the corresponding hexaene (**5**), which in turn was prepared by the sequence: 4-geranyl-2-butyn-1-ol[7] → 4-geranyl-*trans*-2-buten-1-ol ($LiAlH_4$–THF, followed by H_2O)[7] → 1-bromo-4-geranyl-*trans*-2-butene (PBr_3)[8] → **5** ($Ni(CO)_4$ in dimethylformamide, yield 90%),[9,10] purified chromatographically by the thin layer technique (tlc).[11] The 10,15-tritium-labeled hexaene **6** was obtained by a similar sequence with the modification of the reagents in the first step (C≡C reduction) to $LiAlH_4$–$NaOCH_3$, followed by tritium oxide,[7] and the 11,14-tritium-labeled hexaene **7** resulted from an analogous modification with $LiAlH_4$–$AlCl_3$, followed by tritium oxide as reagents for the propargylic reduction.[7] Epoxidation[6] of **6** and **7** afforded the tritiated substrates **8** and **9**. Radiocarbon-labeled oxide **3** was synthesized by conversion of the unlabeled oxide **3** to the trisnoraldehyde (H_3O^+, followed by sodium periodate[1a]) which was transformed to labeled **3** using ^{14}C-labeled diphenylsulfonium isopropylide.[12]

Racemic tritium-labeled oxide **9** (0.275 μmol, specific activity 4×10^6 dpm/μmol) was incubated anaerobically with a solution of 2,3-oxidosqualene–sterol cyclase (60 ml in 0.1 *M* phosphate buffer at pH 7.4) prepared from 25 g of hog liver microsomes[13] for 5 hr at 37°, and the extracted lipid was subjected to two successive tlc separations using silica gel (buffered to pH 7) and 3% ethyl acetate in benzene. In addition to unchanged oxide **9** (R_f 0.65), there was obtained a new labeled product, R_f 0.3, in 38% yield (based on utilization of one antipode of **9**). This material was further purified by the following sequence of operations: (1) tlc using 10% $AgNO_3$ on silica gel and 20% ethyl acetate in chloroform (R_f 0.4); (2) acetylation with acetic anhydride (10 μl) and pyridine (10 μl) at 25° for 16 hr and tlc separation using silica gel–benzene (R_f 0.5); (3) tlc separation using 10% $AgNO_3$–silica gel with 35:65 chloroform–petroleum ether (bp 30–60°) mixture (R_f 0.25). Analysis by gc at this stage using a 10-ft, 0.125-in. column of 2% OV-1 (Supelco, Inc.) silicone on silanized support (Gaschrome Q) at 250° showed the product to have retention time (t_r) (flow rate 60 ml/min) 18.3 min and the only important impurity to be cholestanyl acetate (t_r 21 min).[14] The mass spectrum