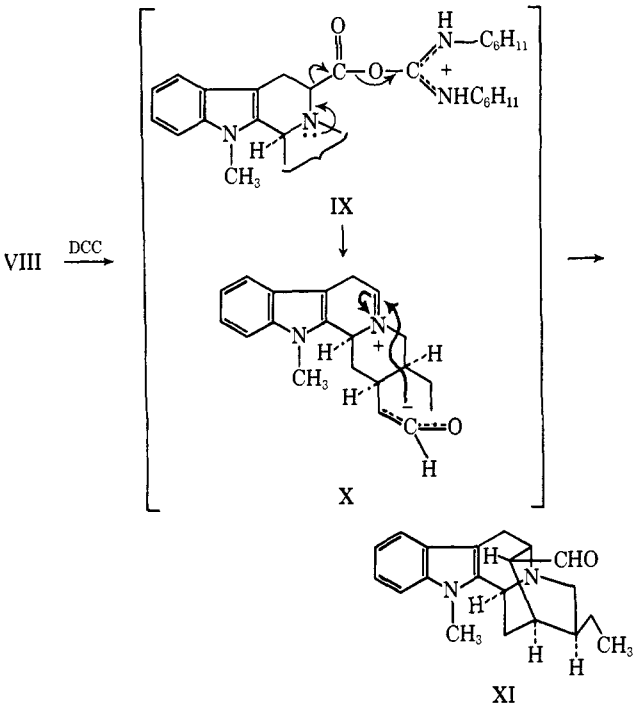
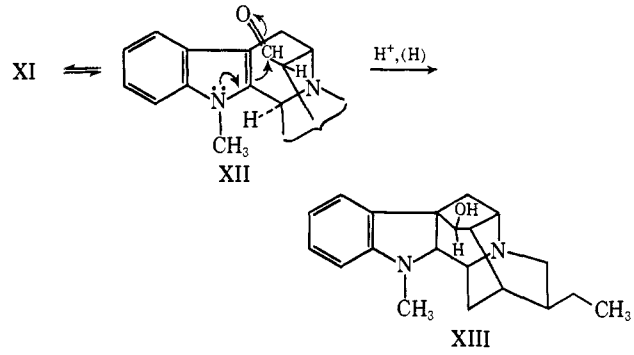
With the carboxyl in VIII properly positioned for directing formation of iminium ion which would permit requisite bond formation between C-5 and C-16,[9] a decarbonylation reaction (IX) was induced by treat-



ment of the amino acid with 2 mol each of dicyclohexylcarbodiimide and *p*-toluenesulfonic acid in dioxane at 80°.[10] The dihydro-β-carboline X was not isolated, but allowed to generate spontaneously, in a bioorganic cyclization (X), *dl*-deoxyajmalal-B (XI) (mp 204–206°), isolated by tlc on silica gel (18%). The synthetic aldehyde was resolved by use of D-camphor-10-sulfonic acid, the resolved base (mp 212–213°) as well as its sulfonate salt (mp 236–240°) being identical with authentic specimens[11] in all respects, including mixture melting points.

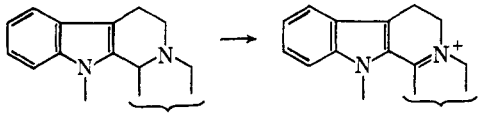
Completion of the synthesis depends on certain relay operations. By means of appropriate experiments carried out with either deoxyajmalal-A (XII) or -B (XI) in room temperature acetic acid–sodium acetate or in refluxing benzene over alumina, it was demonstrated that there exists at equilibrium a mixture of ~15% A and ~85% B (by nmr analysis), from which mixture there can be isolated (tlc, silica gel GF) deoxyajmalal-A (mp 179–180°), identical with authentic base.[11] Reductive cyclization according to the method



of Taylor, *et al.*,[11] brings about biogenetic-type conversion of aldehyde -A (XII), but not -B (XI), to deoxyajmaline (XIII). Functionalization of the latter at C-21 is achieved by the phenyl chloroformate ring opening–oxidative ring closure sequence innovated by Hobson and McCluskey,[12] with resultant formation of ajmaline itself.[13]

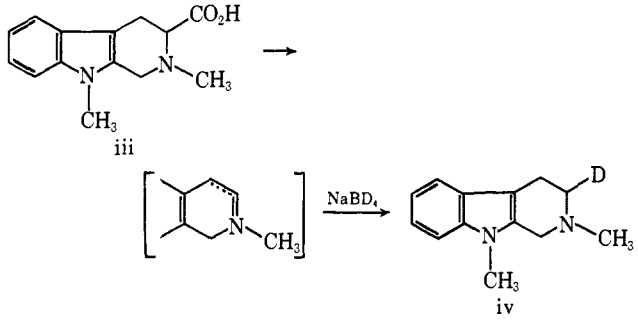
Acknowledgment. The authors are grateful to the National Science Foundation for grant support (GP 7187), and to Dr. M. F. Bartlett, CIBA, for samples of ajmaline and certain of its transformation products.

(9) It is evident that generation of the iminium salt (X) required for the critical C-5–C-16 bond formation would not be possible by dehydrogenation of a tryptamine-derived tetrahydrocarboline (i) with any known reagents (*e.g.*, mercuric acetate), since in such cases the thermody-



namically more stable Δ³-dihydro-β-carboline (ii) results.

(10) The conditions for the decarbonylation reaction were worked out in a model case utilizing the tetrahydro-β-carboline carboxylic acid iii.



Under the conditions used with VIII, the model acid iii was converted to product, reduced *in situ* with $NaBD_4$ to 3-monodeuterio-N-methyl tetrahydro-β-carboline (iv). The latter possessed 60-MHz $CDCl_3$ nmr peaks at *inter alia* τ 6.39 (1 H, doublet, 16 Hz), 5.82 (1 H, doublet, 16 Hz) (C-1 hydrogens), and at 7.00–7.30 (3 H, multiplet) (C-3 and C-4 hydrogens), thus indicating the deuterium site and therefore the nature of the unsaturation in the precursor. For related decarbonylation processes, see V. I. Maksimov, *Tetrahedron*, 21, 687 (1965).

(11) M. F. Bartlett, B. F. Lambert, H. M. Werblood, and W. I. Taylor, *J. Amer. Chem. Soc.*, 85, 475 (1963).

(12) J. D. Hobson and J. G. McCluskey, *J. Chem. Soc.*, 2015 (1967).

(13) Structures of intermediates are supported by all other spectral and analytical data obtained.

(14) National Science Foundation Predoctoral Fellow (1965–1968), National Institutes of Health Predoctoral Fellow (1968–1969).

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Minimal Substrate Structural Requirements for Lanosterol–Squalene 2,3-Oxide Cyclase Action. 10′-Norsqualene 2,3-Oxide

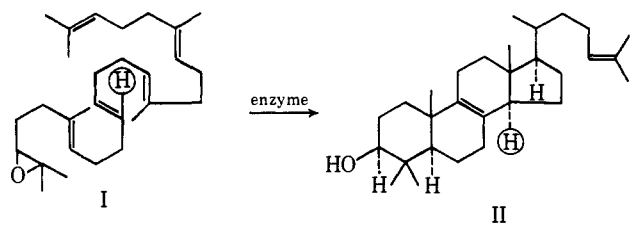
Sir:

One aim of the Stanford investigations into substrate behavior during lanosterol squalene 2,3-oxide cyclase action is definition of the minimal structural requirements for (1) enzymic cyclization and (2) the ensuing methyl–hydrogen migration sequence. The accumulated set of prior preliminary findings has lacked a key case: all-*trans*-10′-norsqualene 2,3-oxide (I). We now wish to report that this oxide—although missing an important methyl group—is converted *in vitro* to 4,4-dimethyl-$\Delta^{8(9),24}$-cholestadienol (II), a

Table I

Oxide	R′, R″	Changing interactions[a]		% conversion
Squalene	CH_3, CH_3	8 → 2 Net decrease	= 6	70–80[b]
15′-Nor-	CH_3, H	5 → 1	4	40–50[b]
10′-Nor-	H, CH_3	3 → 1	2	18–24[b]
10′,15′-Bisnor-[c]	H, H	0 → 0		38% (cyclization only)

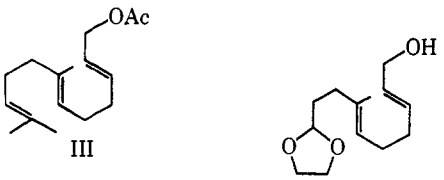
[a] Proto-VI to lanostane system (see text). [b] % conversion of one enantiomer of *d,l*-epoxide to $\Delta^{8(9)}$ sterol under standard conditions. The range for several incubations is given. All material was accounted for by $\Delta^{8(9)}$ sterol, recovered epoxide, and product which was also formed in the boiled enzyme controls. [c] Although this oxide tetracyclized, methyl–hydrogen rearrangement did not then occur; instead side-chain double bond was formed by proton loss: E. J. Corey, P. R. O. de Montellano, and H. Yamamoto, *J. Amer. Chem. Soc.*, **90**, 6254 (1968).



I II

result which, taken together with previous observations, permits *pro tem* specification of the constitutional minima for 1 and 2 above.

Radiolabeled substrate I was prepared along lines previously followed in this laboratory.[1] The *trans,trans*-norfarnesyl acetate III was subjected to the

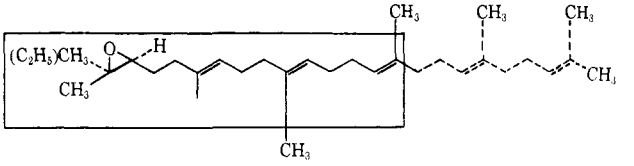


III IV

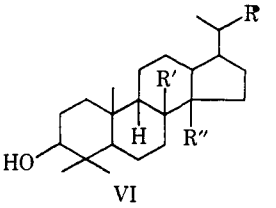
selective terminal oxidation procedure,[2] and the resulting 10,11-oxide was converted to glycol, which was then cleaved to C_{11} aldehyde. The corresponding acetal (IV) was coupled with *trans,trans*-farnesol by means of the Ti(II) intermediated coupling reaction, carried out through the agency of $TiCl_3$–CH_3Li.[3] The resulting all-*trans*-pentaeneacetal, after separation from its congeneric geometrical isomers, was converted to the parent aldehyde, tritium labeled (exchange in acidic ^3H_2O), and then, with diphenylsulfonium isopropylide,[4] transformed to the desired terminal epoxide I.

This substrate was incubated with the cyclase under standard conditions,[1] and for control purposes incubations were carried out with a boiled enzyme preparation. The recovered material was analyzed by tlc, only two bands corresponding to sterol and epoxide being observed. Radioactive material with the same R_f as lanosterol was isolated, trimethylsilated, and analyzed by means of glpc. Roughly 80% of the radioactivity was represented by a peak of R_c 4.05.[5] After W-2 Raney nickel reduction,[6] the enzymic product exhibited the same R_t as authentic dihydro-II. Hydrogenation over platinum[6] gave a product with the same R_t as the $\Delta^{8(14)}$ isomer of dihydro-II. Finally the high-resolution mass spectra of authentic dihydro-II-TMSE and the Raney nickel reduction product (TMSE) were identical, within experimental error.

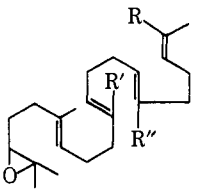
Our extended studies of modified cyclase substrates missing structural elements present in squalene oxide have revealed that cyclization efficiency is not severely diminished by (1) various side chain alternations,[7] (2) absence of the 18-double bond,[8] and (3) absence of the methyl group on C-15.[1] On the other hand, destroying the tertiary center at C-2 by alkyl removal[9] or creating one at C-3 by methyl substitution[10] results in extremely poor cyclization efficiency. Thus, past and present results indicate, in a very general way, that enzymic cyclization may occur when only the sequence framed in formula V is available within some given structure.[11,12] Similarly, in order that *both* cyclization



to a proto skeleton (VI) and ensuing methyl–hydrogen migration take place, the basic system VII must be present.



VI



VII, R′ = CH_3; R″ = H, or R′ = H; R″ = OH

One factor which has recently been stressed in discussions of methyl–hydrogen migrations in triterpenoid rearrangements is the decrease in steric strain and number of nonbonding interactions in the product com-

(1) See, for example, E. E. van Tamelen, R. P. Hanzlik, K. B. Sharpless, R. B. Clayton, W. J. Richter, and A. L. Burlingame, *J. Amer. Chem. Soc.*, **90**, 3284 (1968).

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

(3) K. B. Sharpless, R. P. Hanzlik, and E. E. van Tamelen, *J. Amer. Chem. Soc.*, **90**, 209 (1968).

(4) R. G. Nadeau and R. P. Hanzlik, *Methods Enzymol.*, **15**, 346 (1969).

(5) Retention time relative to cholestane = 1.00 on a 6 ft × 1/8 in. 5% DEGS column at 210°.

(6) F. Gautschi and K. Bloch, *J. Biol. Chem.*, **233**, 1343 (1958).

(7) E. E. van Tamelen, *Accounts Chem. Res.*, **1**, 111 (1968); R. J. Anderson, R. P. Hanzlik, K. B. Sharpless, and R. B. Clayton, *Chem. Commun.*, 53 (1969); J. H. Freed, unpublished results.

(8) E. E. van Tamelen, K. B. Sharpless, R. P. Hanzlik, R. B. Clayton, A. L. Burlingame, and P. C. Wszolek, *J. Amer. Chem. Soc.*, **89**, 7150 (1967).

(9) R. B. Clayton, E. E. van Tamelen, and R. G. Nadeau, *ibid.*, **90**, 820 (1968).

(10) R. P. Hanzlik, unpublished results.

(11) For replacement of oxirane ring methyls by ethyl, see E. E. van Tamelen, R. B. Clayton, and L. O. Crosby, *Chem. Commun.*, 532 (1969).

(12) This generalized representation combines the results of many studies of *individual* modifications and therefore has limited predictive properties.

pared to the reactant.[13] In this connection we note that, although enzymic participation should not be discounted, the results in the table *can* be rationalized without invoking any special enzymic function in directing either the rearrangement or the elimination which terminates it. Focusing attention on only those axial nonbonding interactions (CH_3–CH_3 or H–CH_3) which appear or disappear as the rearrangement of VI occurs, we see in the first three cases that the efficiency as a substrate falls off as the *change* in number of interactions decreases.[14] This trend may be due to removal of an essential part of the driving force for this rearrangement (*i.e.*, relative stabilization of tetracyclic C-20 carbonium ion), thus making the rearrangement step, *at least for the mononor cases*, less facile. The fact that the bisnor compound cyclized efficiently but did not rearrange suggests that the recognition or cyclization steps, *per se*, are not much affected by removal of methyl substituents.[15]

Another important stereochemical change during the rearrangement is that of the B-ring geometry. Evidently, in the absence of both 10′ and 15′ methyls, the strain of the *trans,syn,trans,trans*-ring system alone (as indicated by the bisnor case) does not contribute enough to the driving force to initiate the rearrangement. However, it seems that, with the 10′ and/or 15′ methyls intact, the B boat of the proto system could fortify the driving force, ensuring *continuation to term* ($\Delta^{8(9)}$) of the rearrangement. We wish to emphasize that specifically in the 10′- and 15′-nor cases, no annular double bond isomers were formed by premature elimination. In no case is a Δ^7 isomer found; although concerted loss of a 7β (axial) proton is possible, such elimination would leave a 9β proton, with ring C locked in a boat form. Thus, there is no need to assume that a specific function of the enzyme is removal of the 9β proton; although there may be a specific proton-accepting site in the enzyme, the product is the one which would be formed solely under thermodynamic control.

The above arguments should not be taken to imply that the only function of the cyclizing enzyme is one of protecting reactive intermediates from attack by solvent or bases during the reaction. From our various studies of the nonenzymic-catalyzed cyclization of squalene oxide and other terpenoid epoxides it is obvious that a critical role of the enzyme is specifying a unique chain folding which allows the annelation steps to proceed down *only* one path—a path which is *not* favored thermodynamically.

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(13) P. de Mayo in "Molecular Rearrangements, Part II," P. de Mayo, Ed., Wiley-Interscience, New York, N. Y., 1964, p 821; R. M. Coates, *Tetrahedron Lett.*, 4136 (1967); H. W. Whitlock and M. C. Smith, *ibid.*, 821 (1968); S. C. Pakraski and T. B. Samanto, *ibid.*, 3679 (1967).

(14) Under standard conditions, the yield of product reflects the rate of the overall bimolecular (enzyme + substrate) reaction. This rate in turn is composed of the rates of a number of sequential substeps: (1) formation of E–S complex, (2) tetracyclization, (3) rearrangement to lanostane, and (4) elimination of 9 proton (and separation of enzyme and product molecules at some point).

(15) Linking the C-20 charge center to a conjugated system may also provide stabilization comparable to the removal of both migrating methyls. See E. J. Corey, K. Lin, and H. Yamamoto, *J. Amer. Chem. Soc.*, **91**, 2132 (1969).

(16) National Science Foundation Predoctoral Fellow, 1966–present.

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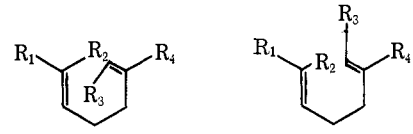
A General 1,5-Diene Synthesis Involving Overall Allyl Alcohol Coupling with Geometrical and Positional Control

Sir:

The general utility and importance of 1,5-dienes with double bonds in specific geometrical arrangements (*e.g.*, juvenile hormones and sterol and cyclic terpene precursors) has made the practical synthesis of pure substances of, for example, types I and II the object of extensive investigation, resulting in the invention or novel development of useful coupling processes.[1-4] However, at the time we initiated this project, there did not exist a general method for performing this operation with the maintenance of the stereochemical and structural integrity of allylic systems essential to a truly catholic procedure. The need for such geometrically pure 1,5-dienes has climaxed in the development of a method for symmetric or asymmetric overall coupling of allyl alcohols which proceeds in good yield with essentially complete preservation of the position and geometry of the olefinic bonds.



I II

R_1, R_2, R_3, R_4 = H or alkyl

In its final form the synthetic sequence involves (1) conversion of the allyl alcohols to allyl bromides by means of carbon tetrabromide and triphenylphosphine, (2) quaternization of tributylphosphine with one of the allyl bromides,[5] (3) C-allylation of the derived ylide with

(1) E. E. van Tamelen and M. A. Schwartz, *J. Amer. Chem. Soc.*, **87**, 3277 (1965); K. B. Sharpless, R. P. Hanzlik, and E. E. van Tamelen, *ibid.*, **90**, 209 (1968).

(2) E. J. Corey and M. F. Semmelhack, *ibid.*, **89**, 2755 (1967), and references cited therein.

(3) G. Stork, P. A. Grieco, and M. Gregson, *Tetrahedron Lett.*, 1393 (1969). The example cited does not allow one to differentiate between coupling at the two possible reactive sites in an allylic Grignard reagent.

(4) J. F. Biellman and J. B. Ducep, *ibid.*, 3707 (1969), describe a sulfur-based coupling procedure terminating with lithium–ethylamine reduction.

(5) It was known[6] that the reaction of triphenylphosphine with primary allylic halides prepared by one of two known stereospecific methods[7,8] yielded primary phosphonium salts with retention of geometrical configuration.