

culations carried out on the 3-21G optimized geometries using a 6-31G* basis and many-body perturbation theory to second order to calculate the electron correlation energy (Table II). However, the agreement between theory and experiment is not as good as the 3-21G level⁵ (Table II) with the relative energy of tautomer 3 predicted to be so high that it would be unobservable in the gas phase.

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Supplementary Material Available: Three tables of observed and calculated transition frequencies used to derive the data in Table I (10 pages). Ordering information is given on any current masthead page.

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Vinyl Group Rearrangement in the Enzymatic Cyclization of Squalenoids: Synthesis of 30-Oxysterols

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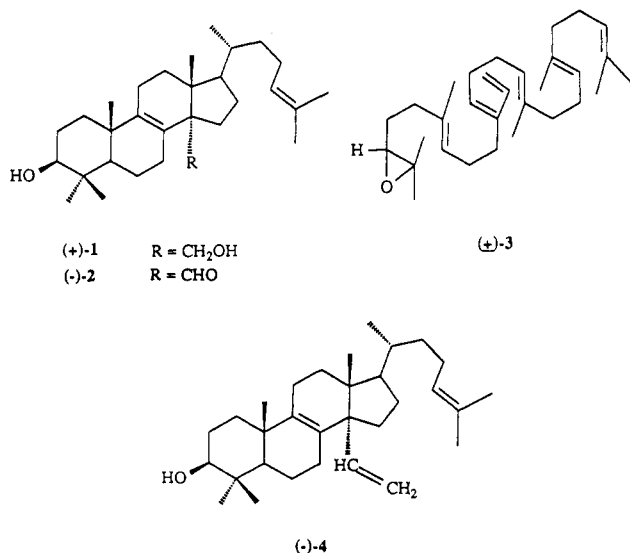
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There is currently an intense search for agents which inhibit HMG-CoA reductase due to their potential use as hypocholesteremic drugs.¹ The tentative identification of 30-hydroxylanosterol (**1**) and its corresponding aldehyde **2** in mevalonate-treated cell cultures suggested that they may be natural receptor-mediated feedback inhibitors of HMG-CoA reductase.² This view is supported by studies which have shown that the 24,25-dihydro derivatives of **1** and **2** strongly suppress HMG-CoA reductase activity³ presumably at the level of transcription that involves binding of the sterol to a specific, intracellular receptor.^{3b,4}

We wish to report the first synthesis of the putative natural oxysterols (+)-**1** and (-)-**2** where the key step involves the enantioselective enzymatic cyclization of the internally functionalized substrate **3** using bakers' yeast to obtain the lanostatriene (-)-**4**. Moreover, the enzymatic conversion of the C-10 vinylic substrate **3** to **4** is the first demonstration of the remarkable ability of the oxidosqualene-lanosterol cyclase to rearrange a substituent other than a hydrogen or methyl group along with the normal sequence of migrations which generates the natural lanosterol skeleton.

The substrate for the enzymatic reaction was constructed by the convergent sequence shown in Scheme I.⁵ The anion from methyl 2-[(bis(trifluoroethyl)phosphono)acetate⁶ was alkylated with homogeranyl iodide⁷ to afford ester **5** which was converted,



via the standard terminal epoxidation procedure,⁸ to the epoxy ester **6**. Oxidation of the farnesylic alcohol **7** to the corresponding aldehyde **8** followed by the stereoselective coupling⁶ of **8** to the anion from ester **6** and KN(TMS)₂/18-crown-6 gave a 58% yield of the α,β -unsaturated ester **9** after chromatographic separation of a 6.5:1 mixture of *Z/E*-isomers. Reduction of **9** using AlH₃¹⁰ in THF afforded the epoxy allylic alcohol **10**¹¹ which was transformed to the aldehyde **11** by PDC¹² in DMF. Lastly, condensation of aldehyde **11** with 1.5 equiv of Ph₃P=CH₂ produced the desired vinylic substrate **3**.

The enzymatic cyclization of the substrate to the desired lanostane intermediate involved the anaerobic incubation of 1.00 g of (\pm)-**3** and 14 g of Triton X-100 with 1.5 L of ultrasonicated bakers' yeast homogenate (150 g of yeast in 0.10 M phosphate buffer prepared as previously described^{13,14}) at 23 °C for 48 h to give, after extractive workup with ether and silica gel chromatography, 0.310 g of (-)-**4** ([α]_D²³ = -47.3°, 62% conversion based on one enantiomer of (\pm)-**3**) as the only new sterol product. Control incubations using a boiled enzyme homogenate failed to produce any new sterol products. Since the ¹H NMR spectrum¹⁶ of **4** displayed chemical shift values for the C-18,19,21,28, and 29 methyls and the C-20 proton (1.44 ppm) consistent with those in lanostane sterols¹⁷ but not the unrearranged dammarane skeleton (C-20-H, 2.42 ppm),^{17,18} we inferred that **4** also possessed the Δ^8 -olefinic bond. This assignment was ultimately confirmed by converting **4** to the known 24,25-dihydro analogues of (+)-**1** and (-)-**2**.

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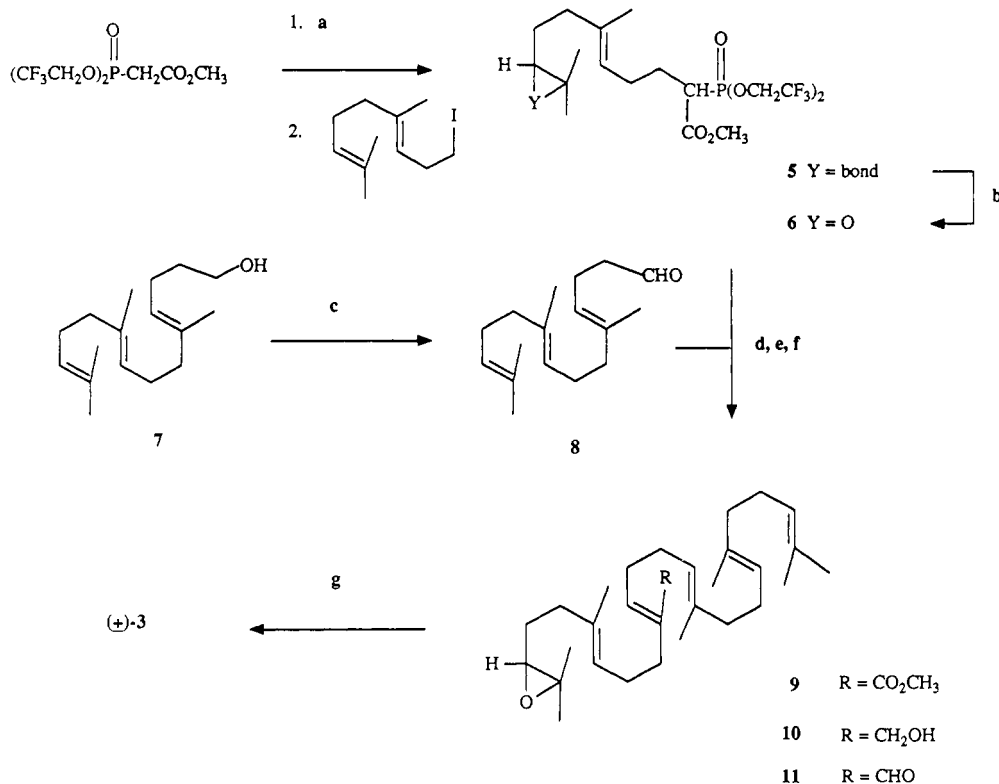
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Scheme 1^a

^a (a) 1.1 equiv of KH, 1 equiv of 18-crown-6, 10% HMPA-THF, -40 °C, 1 h. (b) 1. 1.2 equiv of NBS, 30% aqueous THF, 0 °C, 1 h; 2. K₂CO₃, CF₃CH₂OH (74% yield). (c) 6 equiv of CrO₃, 12 equiv of C₅H₅N, CH₂Cl₂, 0 °C, 6 h (93%). (d) 1.1 equiv of KN(TMS)₂, 5 equiv of 18-crown-6, THF, -50 °C, 1.5 h (58% Z-isomer). (e) 1 equiv of AlH₃, THF, -78 °C, 6 h (68%). (f) 1.5 equiv of pyridinium dichromate, DMF, -15 °C, 2 h (89%). (g) 1.5 equiv of Ph₃P=CH₂, 10% HMPA-THF, -15 °C, 30 min (90%).

The completion of the synthesis of the putative natural sterols was accomplished as follows: **4** was converted to the 3 β -acetate and then treated with 1.2 equiv of *m*-CPBA in CH₂Cl₂ at 23 °C to afford the 24,25-epoxide (93%). Oxidative cleavage of the C-14 vinyl appendage and sequential deoxygenation of the 24,25-epoxy 3 β -acetate in 1:4 CH₂Cl₂-methanol¹⁹ at -78 °C and treatment of the crude ozonolysis mixture with an excess of Zn/AcOH/NaI²⁰ (-78 °C for 1 h then 40 °C for 6 h) to produce 30-oxolanosteryl acetate. Cleavage of the 3 β -acetoxy group by K₂CO₃/MeOH gave 30-oxolanosterol, (-)-**2**, [α]_D²³ = -322°, in 43% overall yield from the lanostatriene (-)-**4**. Lastly, reduction of (-)-**2** with NaBH₄ in methanol at 0 °C gave (+)-30-hydroxylanosterol **1** (98%) [α]_D²³ = +57°. Support for the identity of the synthetic sterols (+)-**1** and (-)-**2** was obtained by hydrogenating (1 atm H₂, PtO₂, 23 °C) each sterol to afford the corresponding 24,25-dihydrosterols whose melting points, IR, NMR, mass spectroscopic, and optical rotation data were in agreement with those previously reported.^{3a,21}

The synthesis described herein illustrates a new approach to the asymmetric preparation of C-30 functionalized lanosterols where the key transformation invokes oxidosqualene cyclase in bakers' yeast for the construction of the steroid nucleus from a completely acyclic progenitor. However, an attempt to apply this enzymic cyclization method to an isomeric substrate possessing a vinyl appendage at C-15 in the squalene backbone was not successful. This latter result supports our recent hypothesis that structural features perturbing the β -face region, but not the α -face,

of the substrate's chair-boat-chair conformation interfere with the enzyme's normal cyclizing operation.¹⁴

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The Furan Approach to Higher Monosaccharides. A Concise Total Synthesis of (+)-KDO

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The higher monosaccharide 3-deoxy-D-manno-2-octulosonic acid, (+)-KDO (**1**), is the essential ketosidic component that links the carbohydrate and lipid subunits of lipopolysaccharides (LPS) of Gram negative bacteria;² incorporation of KDO appears to be vital for the growth and proliferation of these bacteria. Significant interest in the design and synthesis of KDO analogues as potential antibiotics³ has been aroused consequent to recent discoveries that derivatives of 2-deoxy-KDO are effective inhibitors of LPS biosynthesis.⁴ Although several syntheses of KDO and its analogues have been reported,⁵ with one exception,^{5c} carbohydrate precursors

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