crystallize to give a mixture of diastereomeric **12** (mp 211–224 °C dec) in 75% yield.

1,2,3,4,5,6,7,8-Octahydroanthracene-1,5-dicarboxylic Acid. To a solution of 1,2,3,4,5,6,7,8,9,10-decahydroanthracene-1,5-dicarboxylic acid (1.0 g, 3.6 mmol) in anhydrous dioxane (200 mL) was added 2,3-dichloro-5,6-dicyanobenzoquinone (0.9 g, 3.9 mmol). The mixture was refluxed for 2 h, cooled, and filtered. The filtrate was rotary evaporated to give the diastereomeric symmetrical octahydro diacids (mp 220–230 °C dec) in 80% yield after recrystallization from acetone.

Acknowledgment. We thank Marc Rosoff for his help on the "bay side" of phenanthrene.

Registry No. **1**, 82817-89-8; **2**, 82817-90-1; **3**, 82817-91-2; **4**, 110028-98-3; **5**, 59795-49-2; **6**, 113162-64-4; *cis*-**7**, 113162-50-8; *trans*-**7**, 113162-51-9; *cis*-**8**, 113162-65-5; *trans*-**8**, 113162-66-6; *cis*-**9**, 113162-58-6; *trans*-**9**, 113162-59-7; **10**, 38378-77-7; *cis*-**11**, 113162-67-7; *trans*-**11**, 113162-68-8; *cis*-**12**, 113162-52-0; *trans*-**12**, 113162-53-1; **13**, 41694-83-1; **14**, 5462-82-8; *cis*-**15**, 113162-54-2; *trans*-**15**, 113162-55-3; **16**, 113162-69-9; *cis*-**17**, 113162-70-2; *trans*-**17**, 113162-71-3; 1,4,4a,5,8,8a,9,9a,10,10a-decahydro-1,8-anthracenedicarboxylic acid, 113162-72-4; *cis*-1,2,3,4,5,6,7,8-octahydro-1,8-bis[(1-oxoethyl)oxy]anthracene, 113162-73-5; *trans*-1,2,3,4,5,6,7,8-octahydro-1,8-bis[(1-oxoethyl)oxy]anthracene, 113162-74-6; *cis*-1,2,3,4,5,6,7,8-octahydro-1,8-anthracenediol, 113162-60-0; *trans*-1,2,3,4,5,6,7,8-octahydro-1,8-anthracenediol, 113162-61-1; 1,2,3,4,5,6,7,8,9,10-decahydroanthracene-1,5-dicarboxylic acid, 113162-75-7; *cis*-1,2,3,4,5,6,7,8-octahydroanthracene-1,5-dicarboxylic acid, 113162-56-4; *trans*-1,2,3,4,5,6,7,8-octahydroanthracene-1,5-dicarboxylic acid, 113162-57-5; 1,2,3,4,5,6,7,8-octahydro-1,5-bis[(1-oxoethyl)oxy]-anthracene, 113162-76-8; *cis*-1,2,3,4,5,6,7,8-octahydro-4,5-phenanthrenediol, 113162-62-2; *trans*-1,2,3,4,5,6,7,8-octahydro-4,5-phenanthrenediol, 113162-63-3; 5-hydroxy-1,2,3,4,5,6,7,8-octahydro-4-phenanthrenecarboxylic acid, 113162-77-9.

Potential Inhibitors of Vitamin D Metabolism: An Oxa Analogue of Vitamin D[1]

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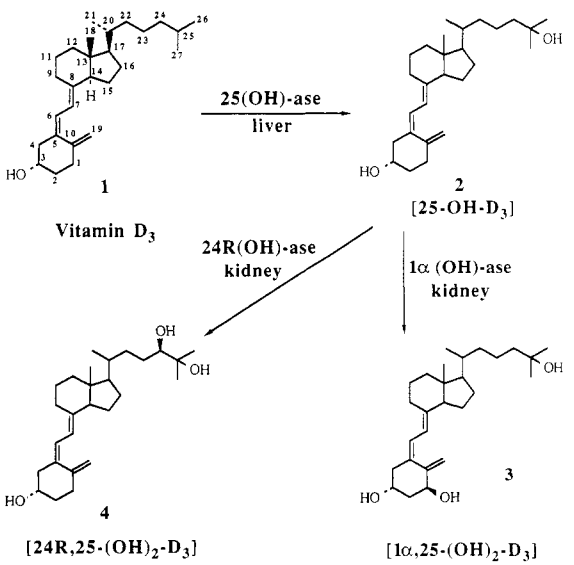
Inhibitors of the enzyme which is responsible for hydroxylation of 25-hydroxyvitamin D_3 (**2**) to the biologically active form 1α,25-dihydroxyvitamin D_3 (**3**) would serve as useful biochemical research tools. The syntheses of 25-hydroxy-3-deoxy-2-oxavitamin D_3 (**5**), a potential inhibitor of 25-hydroxyvitamin D_3 1α-hydroxylase, and the novel seco-A ring vitamin 2-nor-1,3-seco-1,25-dihydroxyvitamin D_3 (**8**) are described. The CD ring ketone **12**, readily prepared via an improved synthetic sequence, was converted to the enol triflate **10**. Coupling of **10** with enyne lactone **27** afforded dienyne lactone **26**. Hydrogenation of **26** produced tetraene **35**, which was oxymercurated–demercurated and then reduced with diisobutylaluminum hydride (DIBAL) to give the novel seco-vitamin **8**. Alternatively, treatment of **35** with DIBAL afforded diol **37**. Cyclodehydration of **37** was achieved by treatment with *n*-BuLi and tosyl chloride to afford **40** and **41**, which after oxymercuration–demercuration gave the desired target molecule **5** and its previtamin form **42**. Vitamin **40** and previtamin **41** (as well as **5** and **42**) are readily interconverted at room temperature via a remarkably facile [1,7]-sigmatropic hydrogen shift. The ratio of vitamin to previtamin for the oxavitamins was determined to be 56 to 44. This is in marked contrast to the behavior of the parent vitamin D_3 system, which exists primarily in the vitamin D rather than the previtamin D form.

Introduction

The principal pathway of vitamin D metabolism (Scheme I) entails three hydroxylations of primary significance.[2,3] The first step involves hydroxylation at the C_{25} position of vitamin D_3 (**1**, D_3) in the liver[4] and the second involves competitive hydroxylation at C_1 and C_{24} of 25-hydroxyvitamin D_3 (**2**, 25-OH-D_3) in the kidney[5] to

Scheme I



(1) This is Paper 34 in the series, Studies of Vitamin D (Calciferol) and Its Analogues. For Paper 33, see: Okamura, W. H.; Hoeger, C. A.; Miller, K. J.; Reischl, W. *J. Am. Chem. Soc.* **1988**, *110*, 973. This article was taken in part from the Ph.D. thesis submitted to the University of California, Riverside, by S. A. Barrack, 1987.

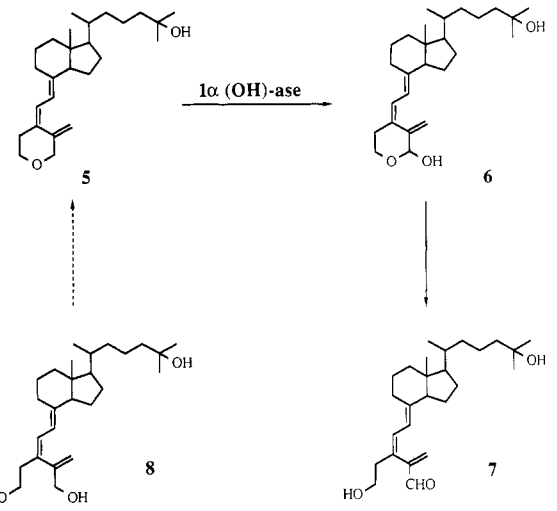
(2) For reviews of the chemistry and biochemistry of Vitamin D, see: (a) Norman, A. W. *Vitamin D the Calcium Homeostatic Hormone*; Academic: New York, 1979. (b) DeLuca, H. F.; Paaren, H. E.; Schnoes, H. K. *Top. Curr. Chem.* **1979**, *83*, 1. (c) Georghiou, P. E. *Chem. Soc. Rev.* **1977**, *6*, 83. (d) Fieser, L. F.; Fieser, M. *Steroids*; Reinhold: New York, 1959. (e) Pardo, R.; Santelli, M. *Bull. Chim. Soc. Fr.* **1985**, 98.

(3) For previous studies of inhibitors of vitamin D_3 25-hydroxylase, see: (a) Johnson, R. L.; Okamura, W. H.; Norman, A. W. *Biochem. Biophys. Res. Commun.* **1975**, *67*, 797. (b) Norman, A. W.; Johnson, R. L.; Okamura, W. H. *J. Biol. Chem.* **1979**, *254*, 11450. (c) Onisko, B. L.; Schnoes, H. K.; DeLuca, H. F. *Tetrahedron Lett.* **1977**, 1107. (d) Onisko, B. L.; Schnoes, H. K.; DeLuca, H. F. *Vitamin D, Basic Research and Its Clinical Application*; Norman, A. W., Shafer, K., Herrath, D. v., Grigolet, H.-G., Coburn, J. W., DeLuca, H. F., Mawer, E. B., Suda, T., Eds.; Walter de Gruyter: Berlin, 1979; p 77. Inhibitors of the 25-OH-D_3-1α-hydroxylase are unknown.

(4) Horsting, M.; DeLuca, H. F. *Biochem. Biophys. Res. Commun.* **1969**, *36*, 251.

0022-3263/88/1953-1790$01.50/0

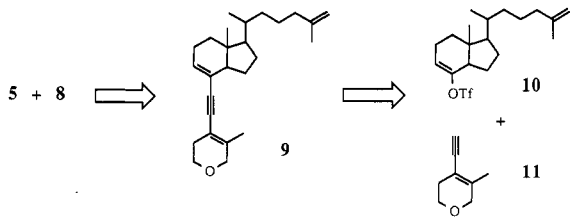
Scheme II



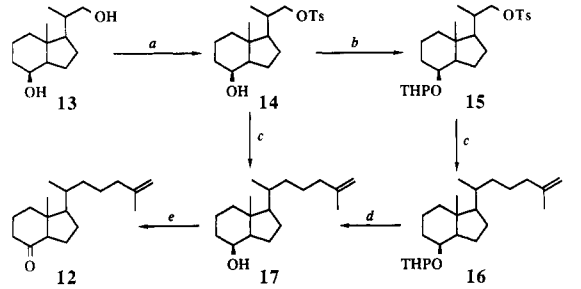
afford 1α,25-dihydroxyvitamin D_3 (**3**, 1α,25-$(OH)_2$-D_3) and 24(*R*),25-dihydroxyvitamin D_3 (**4**, 24(*R*),25-$(OH)_2$-D_3), respectively. The former, **3**, is considered to be the hormonally active form of vitamin D. From the kidney 1α,25-$(OH)_2$-D_3 (**3**) is transported to its target tissues, the intestine, where it is responsible for intestinal calcium absorption (ICA), and the bone, where it stimulates bone calcium mobilization (BCM). In addition to these calcitropic effects (ICA and BCM), this hormone seems to possess more global biological functions. For example, 1α,25-$(OH)_2$-D_3 (**3**) is in some way connected to the normal growth and maturation of certain cells[6] (cell proliferation and differentiation). Thus, analogues of **3** may be clinically useful as drugs in the treatment of certain cancers and skin disorders.[7] The mode of action of 1α,25-$(OH)_2$-D_3 (**3**) for eliciting these physiological effects (ICA, BCM, etc.) is believed to be similar to that of other steroid hormones.[8]

We have recently become particularly interested in the design of inhibitors of the enzyme which converts 25-OH-D_3 (**2**) to its hormonally active form 1α,25-$(OH)_2$-D_3 (**3**).[9] In this regard the A-ring cyclic ether **5** (3-deoxy-25-hydroxy-2-oxavitamin D_3)[10] seems an attractive initial target for serving as a suicide inhibitor[11] in the manner described in Scheme II. Namely, one might envisage the 1α-hydroxylation of **5** to afford the hemi-acetal **6** for which a significant driving force for the ring opening to the α,β-unsaturated aldehyde **7** might exist as a consequence of conjugation. The production of **7** at the active site of 25-OH-D_3-1α-hydroxylase would introduce a functional group possessing exceptionally high Michael addition reactivity, thus offering the possibility of covalent binding to a nucleophilic site on the enzyme.

Scheme III



Scheme IV[a]



[a] (a) TsCl, py (93%); (b) DHP, TsOH (ref 15); (c) $ClMg(CH_2)_2C(CH_3)=CH_2$ (**18**), Li_2CuCl_4, THF (89%); (d) TsOH, EtOH (ref 15); (e) PDC, PTFA, CH_2Cl_2 (82%).

As a precursor to **5**, the novel 1α,25-$(OH)_2$-D_3 analogue **8** was targeted for synthesis.[12] This compound would constitute the first example of a seco A-ring analogue of vitamin D. Such an analogue would be interesting in its own right and might serve usefully as a ligand for developing affinity chromatography systems[13] for purification of the 25-OH-D_3-1α-hydroxylase enzyme as well as the 1α,25-$(OH)_2$-D_3 (**3**) hormone receptor.[14] It is the purpose of this paper to describe the synthesis of the analogues **5** and **8** as well as the unusual effect on the previtamin D/vitamin D equilibrium when an oxygen atom is introduced in the normal A-ring of vitamin D. A significantly improved method for the production of a useful vitamin D CD-ring synthon is also described.

Results and Discussion

The strategy for preparing 25-hydroxy-3-deoxy-2-oxavitamin D_3 (**5**) was to couple the enol triflate **10** with the enyne ether **11** to afford dienyne ether **9** (Scheme III). It was anticipated that Lindlar hydrogenation of **9** followed by [1,7]-sigmatropic hydrogen shift of the resulting triene would afford, after oxymercuration–demercuration, the desired 25-hydroxylated oxavitamin **5**.

Synthesis of the CD Fragment. It was anticipated that the required enol triflate **10** could be readily prepared from the known ketone **12**. Previously, compound **12** had

(5) (a) Frazer, D. A.; Kodicek, E. *Nature (London)* **1970**, *228*, 764. (b) Norman, A. W.; Midgett, R. J.; Myrtle, J. F.; Nowicki, H. G. *Biochem. Biophys. Res. Commun.* **1971**, *42*, 1082. (c) Holick, M. F.; Schnoes, H. K.; DeLuca, H. F.; Gray, W. R.; Boyle, I. T.; Suda, T. *Biochemistry* **1972**, *11*, 4251. (d) Knutson, J. C.; DeLuca, H. F. *Biochemistry* **1974**, *13*, 1543. (e) Henry, H. L.; Taylor, A. N.; Norman, A. W. *J. Nutr.* **1977**, *107*, 1918.

(6) Miyaura, C.; Abe, E.; Kuribayashi, T.; Tanaka, H.; Konno, K.; Nishii, Y.; Suda, T. *Biochem. Biophys. Res. Commun.* **1981**, *102*, 937.

(7) (a) Honma, Y.; Hozumi, M.; Abe, E.; Konno, K.; Fukushima, M.; Hata, S.; Nishii, Y.; DeLuca, H. F.; Suda, T. *Proc. Natl. Acad. Sci. U.S.A.* **1983**, *80*, 201. (b) Koeffler, H. P.; Amatruda, T.; Ikekawa, N.; Kobayashi, Y.; DeLuca, H. F. *Cancer Res.* **1984**, *44*, 5624. (c) MacLaughlin, J. A.; Gange, W.; Taylor, D.; Smith, E.; Holick, M. F. *Proc. Natl. Acad. Sci. U.S.A.* **1985**, *82*, 5409.

(8) Walters, M. R.; Hunziker, W.; Norman, A. W. *Trends. Biochem. Sci.* **1981**, *6*, 268.

(9) This enzyme is believed to belong to the cytochrome P-450 class of oxidase enzymes: (a) Ghazarian, J. G.; Schnoes, H. K.; DeLuca, H. F. *Biochemistry* **1973**, *12*, 2555. (b) Henry, H. L.; Norman, A. W. *J. Biol. Chem.* **1974**, *249*, 7529. (c) Pedersen, J. T.; Ghazarian, J. G.; Orme-Johnson, N. R.; DeLuca, H. F. *J. Biol. Chem.* **1976**, *251*, 3933. (d) For a review of cytochrome P450 enzymes, see: Guengerich, F. P.; Macdonald, T. L. *Acc. Chem. Res.* **1984**, *17*, 9.

(10) There have been three previous reports of oxavitamin D analogues: (a) Toh, H. T.; Okamura, W. H. *J. Org. Chem.* **1983**, *48*, 1414. (b) Kubodera, N.; Miyamoto, K.; Ochi, K.; Matsunga, I. *Chem. Pharm. Bull.* **1986**, *34*, 2286. (c) Murayama, E.; Miyamoto, K.; Kubodera, N.; Mori, T.; Matsunaga, I. *Chem. Pharm. Bull.* **1986**, *34*, 4410.

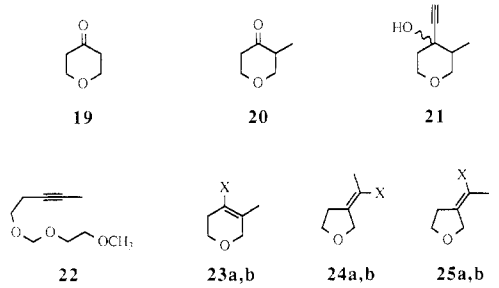
(11) Walsh, C. *Tetrahedron* **1982**, *38*, 871 and references therein.

(12) The analogue **8** is referred to as 2-nor-1,3-seco-1,25-dihydroxyvitamin D_3 in order to more clearly relate its topography and nomenclature to 1α,25-dihydroxyvitamin D_3 (**3**). It is more correctly *A*-nor-3-deoxy-1,2-seco-1,2,25-trihydroxyvitamin D_3.

(13) (a) Parikh, I.; Cuatrecasas, P. *Chem. Eng. News* **1985**, *63* (34), 17. (b) Ross, F. P.; Wecksler, W. R.; Okamura, W. H.; Norman, A. W. *Vitamin D, Basic Research and Its Clinical Application*; Norman, A. W., Shafer, K., Herrath, D. v., Grigolet, H.-G., Coburn, J. W., DeLuca, H. F., Mawer, E. B., Suda, T., Eds.; Walter de Gruyter: Berlin, 1979; p 89, 663.

(14) (a) Norman, A. W.; Roth, J.; Orci, L. *Endocrine Rev.* **1982**, *3*, 331. (b) Wecksler, W. R.; Norman, A. W. *J. Steroid Biochem.* **1980**, *13*, 977.

Chart I

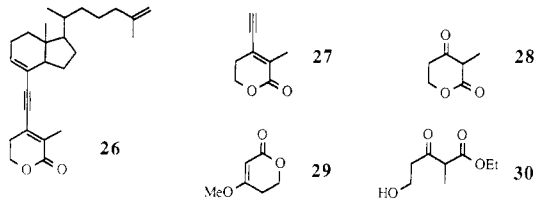


been prepared via the sequence involving **15** and **16** shown in Scheme IV.[15] This route was originally developed by Lythgoe and was later modified slightly by this laboratory to circumvent the partial hydrochlorination of the side chain double bond when **16** is deprotected with aqueous HCl to afford **17**. More recent attempts to prepare larger quantities of **12** via our modified scheme proved cumbersome in part because of the fact that the tetrahydropyranyl (THP) ether protection–deprotection steps were not as efficient as previously thought. Moreover, the presence of THP diastereomers makes purfication inconvenient. We have now found that protection of the 8β-hydroxy functionality is unnecessary;[16] that is, simply by treating tosylate **14** with an excess of the isopentenyl Grignard reagent **18** in the presence of dilithium tetrachlorocuprate, the desired alcohol can be synthesized in excellent yield. The sequence of four steps leading from vitamin D_2 to the diol **13** and then to ketone **12** has been carried out repeatedly in this laboratory. Because of the utility of this ketone in vitamin D analogue synthetic studies, we comment on the reproducibility of each step in some detail in the Experimental Section. Preparation of the desired triflate **10** was achieved in 87% yield by treatment of ketone **12** with lithium diisopropylamide (LDA) to form the kinetic enolate followed by quenching with *N*-phenyl-*N*-[(trifluoromethyl)sulfonyl]trifluoromethanesulfonamide.[17]

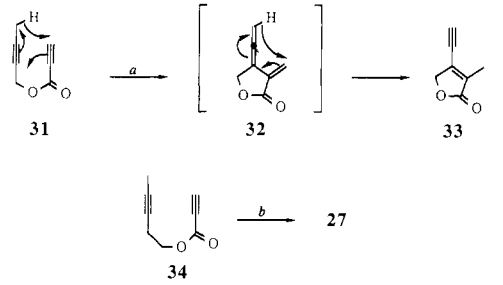
Synthesis of the A-Ring. A simple, logical precursor to enyne **11** was commercially available tetrahydro-4*H*-pyran-4-one (**19**) (Chart I). All attempts to dehydrate the propargyl alcohol **21**[18] derived from the methyl derivative **20**[19] of **19** proceeded very poorly and this straightforward approach had to be abandoned early in this investigation.

An alternative approach to **11** involved construction of the A-ring by means of direct cyclization of an acyclic precursor such as **22** to give **23a** or **23b**. The cyclization approach to compounds of type **23** was based on the work of Thompson,[20] in which tetrahydropyrans were synthesized via the titanium tetrahalide promoted cyclization of unsaturated acetals. By analogy, if the unsaturated acetal contained a triple bond (as in **22**) instead of a double bond, the product should be a vinyl bromide, which could be coupled with an appropriate acetylene moiety to give the desired enyne. Unfortunately, the titanium tetrabromide promoted cyclization of **22**[21] yielded primarily a mixture of the five-membered ring vinyl bromides **24a** and **25a**, with only small amounts of the desired A-ring fragment **23a**. Similarly, triflic acid catalyzed cyclization of acetal **22** also gave primarily **24b** and **25b**, with only a small amount of the desired triflate **23b**.[22] Johnson, in his polyene cyclization studies, has shown that in certain instances, cyclization of related acetylenes to cyclohexenyl rather than methylenecyclopentane systems can be achieved.[24] However, this was not the case here.

Chart II



Scheme V[a]



[a] (a) FVP (flash vacuum pyrolysis), 550 °C [66% (this laboratory); 85%[27]]; (b) FVP, 570 °C (11%).

A slightly different route to **5** and also **8** was devised in which the immediate precursor targeted for the synthesis was the dienyne lactone **26** (Chart II) rather than the ether **9**. We anticipated that the coupling of the A-ring lactone **27** with the enol triflate **10** would be feasible.[17] For synthesizing lactone **27** we envisaged the cyclic β-keto ester **28** as the appropriate precursor. Unfortunately, although precursors such as **29**[25] and **30**[26] could be synthesized, attempts at their conversion to **28** were unsuccessful.

The solution to the synthesis of the desired lactone **27** proved in the end to be quite simple. During the course of our investigations, Dreiding[27] described a most unusual

(15) (a) Lythgoe, B.; Roberts, D. A.; Waterhouse, I. *J. Chem. Soc., Perkin Trans. 1* **1977**, 2608. (b) Leyes, G. A.; Okamura, W. H. *J. Am. Chem. Soc.* **1982**, *104*, 6099. (c) Leyes, G. A. Ph.D. Dissertation, University of California, Riverside, CA, 1981.

(16) After this work was completed, a similar observation was reported: Baggiolini, E. G.; Iacobelli, J. A.; Hennessy, B. M.; Batcho, A. D.; Sereno, J. F.; Uskokovic, M. R. *J. Org. Chem.* **1986**, *51*, 3098.

(17) For related procedures, see: (a) Castedo, L.; Mourño, A.; Sarandeses, L. A. *Tetrahedron Lett.* **1986**, *27*, 1523. (b) Cacchi, S.; Morera, E.; Ortar, G. *Synthesis*, **1986**, 320.

(18) Propargyl alcohol **21** was prepared in 75% yield by the addition of lithium acetylide to ketone **20**: (a) Midland, M. M. *J. Org. Chem.* **1975**, *40*, 2250. (b) Midland, M. M.; McLoughlin, J. I.; Werley, R. T., Jr. *Org. Synth.*, in press.

(19) Ketone **20** was prepared via the classical Stork enamine procedure: Stork, G.; Brizzolara, A.; Landesman, H.; Szmuszkovicz, J.; Terrell, R. *J. Am. Chem. Soc.* **1963**, *85*, 207. Attempts at alkylation of **19** by direct deprotonation with LDA followed by quenching with methyl iodide resulted in a complex mixture of products.

(20) Winstead, R. C.; Simpson, T. H.; Lock, G. A.; Schiavelli, M. D.; Thompson, D. W. *J. Org. Chem.* **1986**, *51*, 275.

(21) Ether **22** was prepared in 92% yield from 3-pentyn-1-ol and MEM-Cl: Corey, E. J.; Gras, J.-L.; Ulrich, P. *Tetrahedron Lett.* **1976**, 809.

(22) Attempts were also made to prepare the thermodynamic enol triflate **23b** from ketone **20**[23] but these led only to recovered starting material. In parallel control experiments, the thermodynamic enol triflate of 2-methylcyclohexanone was formed quite readily.

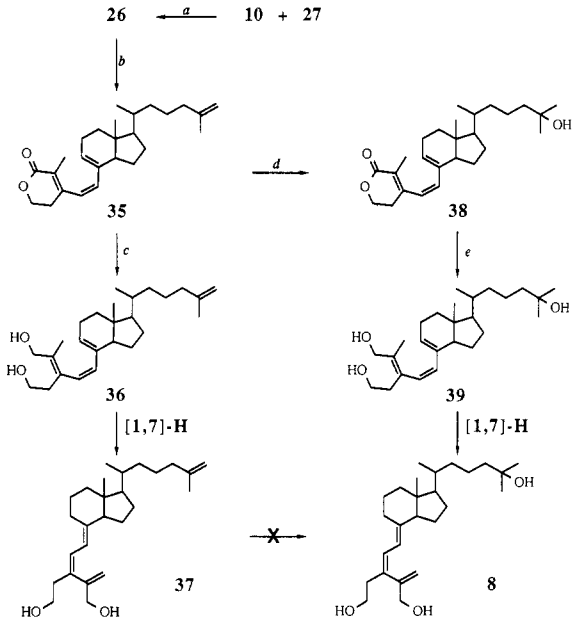
(23) (a) Kraft, M. E.; Holton, R. A. *Tetrahedron Lett.* **1983**, *24*, 1345. (b) McMurray, J. E.; Scott, W. J. *Tetrahedron Lett.* **1983**, *24*, 979.

(24) (a) Johnson, W. S. *Bioorg. Chem.* **1976**, *5*, 51. (b) Johnson, W. S.; Gravestock, M. B.; Parry, R. J.; Okorie, D. A. *J. Am. Chem. Soc.* **1972**, *94*, 8604.

(25) (a) Savard, J.; Brassard, P. *Tetrahedron Lett.* **1979**, 4911. (b) Castellino, S.; Sims, J. J. *Tetrahedron Lett.* **1984**, *25*, 2307. (c) Midland, M. M.; Graham, R. S. *J. Am. Chem. Soc.* **1984**, *106*, 4294. (d) Danishefsky, S.; Harvey, D. F.; Quallich, G.; Uang, B. J. *J. Org. Chem.* **1984**, *49*, 392.

(26) Hannick, S. M.; Kishi, Y. *J. Org. Chem.* **1983**, *48*, 3833.

Scheme VI[a]



[a] (a) $Pd(PPh_3)_2Cl_2$, Et_3N, DMF (81%); (b) Lindlar catalyst, H_2, hexanes (68%); (c) DIBAL, PhH (83%); (d) $Hg(OAc)_2$, THF/H_2O; $NaBH_4$, NaOH, H_2O (67%); (e) DIBAL, PhH (67%).

process which entailed the remarkable rearrangement under flash vacuum pyrolysis (FVP) conditions of the diyne ester **31** to afford lactone **33**, a substance structurally similar to our desired target **27** (Scheme V). The transformation of **31** to **33** apparently proceeds via an initial ene reaction to afford vinylallene **32**, which undergoes a subsequent [1,5]-sigmatropic shift to afford **33**. This remarkable rearrangement reportedly proceeds to afford **33** in 85% yield.

As an extension of this process to our problem, we envisaged the isomerization of the homologous precursor **34**. After a variety of attempts, flash vacuum pyrolysis of diyne ester **34** afforded **27**, but in only 11% yield. In order to determine whether the low yield of **27** obtained in this study was an artifact of our FVP technique, we repeated the synthesis of **33** reported by Dreiding.[27] In our first attempt, pyrolysis of the known **31** (prepared in an improved yield) afforded **33** in 66% yield. Thus we conclude that the low yield of **27** is intrinsic to the course of rearrangement of **34**. We speculate that perhaps **27** is capable of retro-Diels–Alder fragmentation, but this point was not investigated further. Despite the low yields, gram quantities of the desired lactone are easily available and in fact, the precursor **34** can be obtained in only one step from commercial materials. Accordingly, we terminated further efforts to develop alternatives to the deceptively simple **27** or related materials. We were delighted with the fact that **27** could now be obtained in amounts more than necessary to complete the synthesis of **5** and **8**.

Synthesis of 5 and 8. Enol triflate **10** and enyne lactone **27** were coupled[17] in the presence of bis(triphenylphosphine)palladium(II) chloride to afford the desired dienyne lactone **26** in 81% yield (Scheme VI). Hydrogenation of **26** using Lindlar catalyst gave tetraene **35** in an average yield of 60% (seven runs). A side product was formed in this reaction from the over reduction of the acetylene, but this could be minimized by monitoring the reaction by ^1H NMR. Tetraene lactone **35** was treated with 3 equiv of diisobutylaluminum hydride[28] (DIBAL), which afforded a mixture of previtamin **36** and vitamin **37** diols in which the A ring had been opened. The [1,7]-sigmatropic shift of the previtamin **36** to vitamin **37** occured readily at ambient temperature. Interestingly the mass spectrum of diol **37** exhibited m/z 140 (attributed to the A-ring portion by $C_{7,8}$ cleavage) as the base peak. The mass spectrum of vitamin D_3 is uniquely characterized by the appearance of a base peak resulting from a similar $C_{7,8}$ cleavage.[29] Thus, although the diol olefin has no A-ring, the triene moiety in the seco-vitamin **37** behaves similarly to the vitamin D system under electron impact conditions.

Attempts to oxymercurate–demercurate diol **37** to the corresponding triol **8** failed. With the use of 1 equiv of mercuric acetate, only starting material was recovered. Addition of excess mercuric acetate resulted in what appeared to be partial reaction of the conjugated triene portion of **37** rather than the seemingly more nucleophilic side chain double bond. This problem was circumvented by carrying out the oxymercuration at an earlier stage in the synthesis. Lactone **35** was treated with mercuric acetate and then cautiously demercurated with basic sodium borohydride (so as not to saponify the lactone) to produce the hydroxylated A-ring lactone **38** in 67% yield and an essentially quantitative recovery of unreacted starting material. Treatment of the hydroxy lactone **38** with 3 equiv of DIBAL resulted in a mixture of triols **8** and **39**. As noted previously for the diol olefin **36**, which readily isomerizes to **37**, the previtamin triol **39** quantitatively rearranges to vitamin **8** at room temperature on standing for several hours. The mass spectrum of **8**, like that of **37**, also exhibited a base peak at m/z 140 in its mass spectrum.

A variety of methods were attempted to cyclodehydrate the diol **37** to **40**, which was expected to be convertible to the final target **5**. Since the formation of cyclic ethers under basic conditions from related diols by the in situ intramolecular displacement of a tosylate is well precedented in the literature, diol **37** was heated in the presence of tosyl chloride in pyridine. Only starting material could be recovered under the previously reported[30] conditions. In all instances, acidic conditions caused deterioration of the triene moiety. After considerable experimentation, it became readily apparent that the Williamson ether cyclization of **37** would not be as simple as we had anticipated. This may perhaps be a consequence of the presence of two sp^2 centers in the oxa A-ring, resulting in some degree of ring strain.

A mixture of the two possible monotosylates could be prepared by treatment of **37** with 1 equiv of *n*-butyllithium followed by quenching with tosyl chloride, but significant amounts of starting material were usually recovered. In separate experiments, the tosylates, after treatment with *n*-butyllithium, could each be cyclized to **40** by heating in DMF. However, a more efficacious route to the cyclic ethers was to perform the reaction in "one pot" by treating diol **37** with excess in *n*-BuLi in THF, quenching with tosyl chloride, replacing the solvent with DMF, and heating for 12 h (Scheme VII). With this method, the cyclic ethers (a mixture of previtamin **41** and vitamin **40**) were produced in 45% yield (62% yield based on recovered **37**, which could be recycled). The reaction was very clean by TLC, showing only starting material and products.

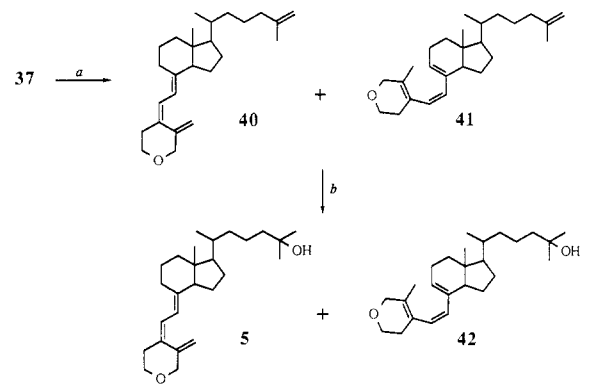
(27) Bilinski, V.; Karpf, M.; Dreiding, A. S. *Helv. Chim. Acta* **1986**, *69*, 1734.

(28) Kreft, A. *Tetrahedron Lett.* **1977**, 1959.

(29) (a) Blunt, J. W.; DeLuca, H. F.; Schnoes, H. K. *Biochemistry* **1968**, *7*, 3317. (b) Okamura, W. H.; Hammond, M. L.; Jacobs, H. J. C.; van Thuijl, J. *Tetrahedron Lett.* **1976**, 4807.

(30) Engel, Ch. R.; Chowdhury, M. N. R. *Tetrahedron Lett.* **1968**, 2107.

Scheme VII[a]



[a] (a) *n*-BuLi, THF; *p*-TsCl; DMF, 60 °C (45%); (b) $Hg(OAc)_2$, THF/H_2O; $NaBH_4$, NaOH, MeOH (22%; 50% based on recovered starting material).

Separation of the previtamin **41** and vitamin **40** proved to be inconvenient (although separation could be achieved using two recycles on HPLC) due to the ready equilibration of vitamin and previtamin. To discern information about this two component mixture and confirm the assignment of signals due to each component, an experiment utilizing the HPLC–NMR[31] technique was employed. Interfacing the flow from the HPLC directly into an observed volume in the probe of a 300-MHz ^1H NMR spectrometer showed that previtamin **41** eluted first, as seen clearly by the angular methyl group signal at δ 0.71. Vitamin **40** eluted second, as seen by its angular methyl group signal at δ 0.56.

The cyclic ethers **40** and **41** could be hydroxylated at C_{25} by treatment with mercuric acetate followed by demercuration with sodium borohydride in aqueous sodium hydroxide. This reaction unfortunately appeared to be very sluggish, resulting in only a 22% absolute yield of the equilibrium mixture of **5** and **42** (a 50% yield based on recovered starting material, which could then be recycled). Although the conversion is low, $NaBH_4$ demercuration provides a means of isotopic labeling (e.g., tritium) in the final synthetic step.

It is known that for vitamin D_3 (**1**) and previtamin D_3 (which possess six-membered carbocyclic A-rings), the equilibrium lies on the side of the vitamin **1** with an equilibrium ratio on the order of 90:10.[32] For an analogue of vitamin D_3 and previtamin D_3 possessing a seven membered A-ring,[33] the equilibrium appears to lie completely on the side of the vitamin. In a recent study from this laboratory (J. Enas, unpublished results), an analogue of vitamin D_3 possessing a five-membered A-ring was found to exist mainly in the previtamin form. Thus, it is interesting that for the 2-oxa analogue, the equilibrium vitamin to previtamin (**40**:**41**) ratio was determined to be 56:44 (^1H NMR integration). This finding is explicable on the basis that because the carbon–oxygen bond length (1.43 Å) is shorter than the carbon–carbon bond length (1.54 Å), the six-membered cyclic ether system **40** and **41** is somewhat smaller than the typical carbocyclic six membered ring. Thus we can tentatively hypothesize that as the ring size decreases, the equilibrium ratio of vitamin to previtamin also decreases. The origin of this effect would appear to be related to the finding that there is greater strain energy in a methylenecyclopentane system than in a methylenecyclohexane system when compared to their corresponding endocyclic counterparts.[34] What remains inexplicable is why interconversion between **40** and **41** appears to be faster (occuring readily at room temperature) than that between vitamin D_3 and previtamin D_3. The latter equilibrium usually requires several hours of heating at >60 °C.

Since the desired target molecule **5** exists to a substantial degree in its tautomeric previtamin form **42**, the question arises as to whether this new analogue mixture will serve effectively as a substrate for its biological receptor. In view of the relative ease with which **42** reverts via a [1,7]-sigmatropic shift to **5** even at room temperature, **42** should be readily siphoned off as **5** if introduced to a receptor such as the enzyme, 25-hydroxyvitamin D_3-1α-hydroxylase. Since this enzyme utilizes 25-OH-D_3 (**2**) as its natural substrate, one can also ask the question whether **5**, which lacks a 3β-hydroxyl group, will be recognized by this enzyme. The finding that 3-deoxy-25-hydroxyvitamin D_3 (i.e., the analogue **5** with the oxygen replaced by a methylene group) exhibits calcitropic activity,[35] implies that the 3-hydroxyl group is not mandatory for 1α-hydroxylation. However, it remains for direct biological evaluation of **5** to determine its efficacy as a inhibitor of the 1α-hydroxylase enzyme. Such studies are currently under way in the laboratories of Professor Helen H. Henry and Professor Anthony W. Norman in the Department of Biochemistry at UC Riverside.

Experimental Section

General. Spectral data are given in the supplementary material. Air-sensitive compounds were generally stored under nitrogen in a −80 °C freezer, and reactions involving organometallic materials were performed under an atmosphere of dry nitrogen or argon. Dry ether or THF (tetrahydrofuran) refers to reagent grade material freshly distilled from sodium–benzophenone ketyl or $LiAlH_4$ under nitrogen. Skellysolve B (hexanes) was distilled from CaH_2 prior to use. Kugelrohr distillation boiling points (bp) refer to the external air bath temperature. High-pressure liquid chromatography (HPLC) was performed with a Waters 6000A solvent delivery system equipped with a U6K injector and dual-detector system (M450 variable wavelength UV and R401 refractive index detectors). A Whatman M9 10/50 Partisil (10-μm particle size, 9.4 mm i.d. × 50 cm) column was used except as noted. All chromatography solvents were distilled prior to use. Solvents and solvent mixtures for HPLC were vacuum-filtered through a 0.45-μm Millipore filter and vacuum degassed immediately prior to use. Flash chromatography was performed with Sigma silica gel (230–400 mesh). Chromatotron refers to preparative, centrifugally accelerated, radial, thin-layer chromatography with silica gel PF-254 with $CaSO_4 \cdot {}^1/_2H_2O$ as the absorbent. Thin-layer chromatography (TLC) was performed with precoated plastic silica gel plates (0.25 mm, Brinkmann). Compounds were detected by UV light or by phosphomolybdic acid–ethanol spray.

De-*A*,*B*-23,24-dinorcholane-8β,22-diol (13, the Inhoffen/Lythgoe Diol). The diol **13** was prepared by ozonolysis of vitamin D_2 according to the procedure develeoped by Castedo and coworkers.[36] In some 30 trials usually on an 8-g scale the yields obtained varied from 40% to 74%, with an average yield of 68%.[37]

(31) Laude, D. A., Jr.; Wilkins, C. L. *Anal. Chem.* **1984**, *56*, 2471.
(32) (a) Hanewald, K. H.; Rappoldt, M. P.; Roborgh, J. R. *Recl. Trav. Chim. Pays-Bas* **1961**, *80*, 1003. (b) For a recent mechanistic study on the [1,7]-sigmatropic hydrogen shift with leading references, see: Hoeger, C. A.; Johnston, A. D.; Okamura, W. H. *J. Am. Chem. Soc.* **1987**, *109*, 4690. See also ref 1.
(33) Sine, S. M.; Conklin, T. E.; Okamura, W. H. *J. Org. Chem.* **1974**, *39*, 3797.
(34) Wiberg, K. B.; Wasserman, D. J.; Martin, E. J.; Murcko, M. A. *J. Am. Chem. Soc.* **1985**, *107*, 6019.
(35) Edelstein, S.; Sheves, M.; Mazur, Y.; Bar, A.; Hurwitz, S. *FEBS Lett.* **1979**, *97*, 241.
(36) (a) Sardina, F. J.; Mouriño, A.; Castedo, L. *J. Org. Chem.* **1986**, *51*, 1264. The diol **13** has also been prepared via total synthesis: (b) Trost, B. M.; Bernstein, P. R.; Funfschilling, P. C. *J. Am. Chem. Soc.* **1979**, *101*, 4378. (c) Wovkulich, P. M.; Barcelos, F.; Batcho, A. D.; Sereno, J. F.; Baggiolini, E. G.; Hennessy, B. M.; Uskokovic, M. R. *Tetrahedron* **1984**, *40*, 2283. (d) Johnson, W. S.; Elliot, J. D.; Hanson, G. J. *J. Am. Chem. Soc.* **1984**, *106*, 1138.

De-A,B-23,24-dinor-22-(p-tolylsulfonoxy)cholan-8β-ol (14). Tosylate **14** was prepared from diol **13** by the method of Leyes[15b,c] with the modifications of Castedo and co-workers.[36a]. The substance was originally reported by Lythgoe as a liquid[15a] and later by Leyes as a crystalline solid (mp 93–95 °C).[15c] The yields obtained ranged from 71% to 96% for 12 runs with an average yield of 86%.

De-A,B-25-cholesten-8β-ol (17). Magnesium turnings (1.32 g, 54.4 mmol), 4-chloro-2-methyl-1-butene[38] (3.06 mL, 27.2 mmol), iodine (2 crystals), and THF (18 mL) were placed in a dry 100-mL flask equipped with magnetic stir bar, West condenser, and side arm with a Teflon stopcock. After the mixture was refluxed for 12 h, the solution was cooled to room temperature and then added dropwise via a 20-gauge cannula to a solution of tosylate **14** (2.00 g, 5.46 mmol) in THF (16.4 mL) at −78 °C. A mixture of LiCl (0.232 g, 5.46 mmol) and $CuCl_2$ (0.368 g, 2.73 mmol) was dried in a 50-mL flask under vacuum at ~50 °C for several hours. After cooling, THF (27 mL) was added to give a 0.1 M solution of Li_2CuCl_4. This solution was then added dropwise via cannula to the stirred mixture of tosylate and Grignard reagent at −78 °C. The cooling bath was then removed and the reaction mixture stirred at room temperature for 10 h and then poured cautiously into a stirred mixture of 150 mL of ice and 50 mL of 1 M H_2SO_4. After the mixture was extracted with ether (3 × 50 mL), the combined organic layers were washed with aqueous NH_4Cl, aqueous $NaHCO_3$, and brine (1 × 50 mL each), dried ($MgSO_4$), filtered, and concentrated. Purification of the residue by flash chromatography (10% EtOAc/hexanes, 15 × 4 cm silica gel column) followed by vacuum-drying gave 1.34 g (93%) of alcohol **17** as a viscous colorless liquid. In eight trials, the yields obtained ranged from 82% to 93%, with an average yield of 89%.

De-A,B-25-cholesten-8-one (12).[15] The alcohol **17** (1.84 g, 6.96 mmol) was dissolved in dichloromethane (5 mL, distilled from CaH_2) and then added via cannula (with 2-mL washing) to a magnetically stirred suspension of pyridinium dichromate (7.86 g, 20.9 mmol) and pyridinium trifluoroacetate (0.537 g, 2.78 mmol) in dichloromethane (18 mL) under N_2. The reaction mixture was then stirred for 4.5 h at room temperature. The resulting black mixture was passed through a fritted glass funnel containing a slurry of diatomaceous earth (CH_2Cl_2) covered by a slurry of silica gel (CH_2Cl_2). The solvent was removed to give a brown oil, which was purified via flash chromatography (5% EtOAc/hexanes, 18 × 5 cm silica gel) to give 1.63 g (89%) of pure ketone **12** as a viscous oil. In seven runs in this laboratory, the yields obtained varied from 76% to 89% with a average yield of 82%. Epimerization of the trans ketone to the cis isomer (readily detected by its ^1H NMR C_{18}-CH_3 signal at δ 1.04)[39] is an occasional problem on attempted distillation of this material. It is best used as a chromatographically pure, vacuum-dried material.

De-A,B-cholesta-8,25-dien-8-yl Trifluoromethanesulfonate (10). Lithium diisopropylamide was prepared by treating diisoproplyamine (0.63 mL, 4.5 mmol) with *n*-BuLi (2.98 mL, 4.5 mmol, 1.60 M in hexanes) in 5 mL of THF at 0 °C. After being stirred for 15 min, the solution was cooled to −78 °C and ketone **12** (1.0 g, 3.8 mmol) in 1 mL of THF (with 2 × 0.5 mL additional rinsings) was added dropwise via cannula. After being stirred for 15 min the enolate solution was warmed to room temperature over 1 h 45 min and then recooled to −78 °C. *N*-Phenyl-*N*-[(trifluoromethyl)sulfonyl]trifluoromethanesulfonamide (1.5 g, 4.2 mmol) was dissolved in 2 mL of THF and added to the enolate at −78 °C. The reaction solution was transferred to a bath maintained at 0 °C, and stirring was continued for 15 h. The solution was transferred to a separatory funnel and washed successively with 10% H_2SO_4, H_2O, 10% NaOH, H_2O, and brine. The extract was dried with $MgSO_4$, filtered, and concentrated. Purification was achieved by passing the residue through a short pad of silica gel and then subsequent chromatography (100% hexanes) to afford 1.3 g (87%) of the pure triflate **10**.

3-Pentyn-1-yl Propiolate (34). Propiolic acid (10.0 g, 143 mmol) and 3-pentyn-1-ol (12.0 g, 143 mmol) were placed in a flask with 90 mL of benzene. After addition of *p*-toluenesulfonic acid (572 mg, 3 mmol) the flask was equipped with a Dean–Stark apparatus, and the resulting solution was heated for 12 h. Workup consisted of dilution with ether followed by washing the organic layer with saturated $NaHCO_3$ and brine. Drying over $MgSO_4$ and removal of solvent under reduced pressure afforded the desired ester. Kugelrohr distillation [bp 60 °C (1 mm)] gave 16.7 g (87%) of pure 3-pentyn-1-yl propiolate (**34**).

Flash Vacuum Pyrolysis Apparatus. The flash vacuum pyrolysis (FVP) apparatus consisted of a quartz chip packed hot tube (vertically mounted quartz tube; hot zone = 40 × 2.5 cm o.d.). The tube was wrapped with nichrome wire (the length and gauge was adjusted to allow for heating the tube up to 600 °C at the full Variac setting) and then overlayed with a thick layer of asbestos for insulation. A cylindrical outer glass shield was clamped around the hot zone to provide further insulation. The quartz tube possessed an internal well for inserting a thermocouple to monitor the temperature. Attached to the top of the quartz tube was an adapter connected to a flask equipped with a side arm (used as a nitrogen and/or sample inlet). The flask could be immersed in a heated oil bath so that substrate could be introduced as a vapor under a nitrogen stream into the hot tube. Attached to the bottom of the hot tube were a series of traps (cooled with liquid N_2) all connected to a vacuum pump.

3-Ethynyl-2-methyl-6-oxacyclohex-2-en-1-one (27). The diynolate **34** (2.20 g, 16.2 mmol) was distilled (oil bath at 120 °C) into the FVP quartz tube according to the procedure given in the preceding section. A slow flow of N_2 (30 mL/min) facilitated directing the vapors into the hot tube, which was heated to 570 °C at approximately 30 Torr of pressure. The vapors were condensed under vacuum into three traps connected in series and each cooled with liquid N_2 as described. The hot tube and distillation flask were allowed to cool to room temperature, then the traps were removed, and the system was brought to ambient pressure. After rinsing of all of the traps with ether and then evaporation the ether solution to dryness, the crude product was passed through a short silica gel column (25% EtOAc/hexanes). The concentrated residue was recrystallized from an ether–hexanes mixture to afford crystalline **27** (250 mg, 1.8 mmol, 11%, mp 89–90 °C).

2-Butyn-1-yl Propiolate (31). Propiolic acid (4.0 g, 57.1 mmol) and 2-butyn-1-ol (4.0 g, 57.1 mmol) were placed in a flask with 40 mL of benzene. After addition of *p*-toluenesulfonic acid (213 mg, 1.1 mmol), the flask was equipped with a Dean–Stark apparatus, and the resulting solution was heated for 12 h. Workup consisted of dilution with ether followed by washing with saturated aqueous $NaHCO_3$, H_2O, and brine. Drying over $MgSO_4$ and removal of solvent under reduced pressure afforded the desired ester. Kugelrohr distillation [bp 114 °C (2 mm)] gave 5.8 g (83%) of pure 2-butyn-1-yl propiolate (**31**).

3-Ethynyl-2-methyl-5-oxacyclopent-2-en-1-one (33). As in the preparation of **27**, Dreiding's diynolate[27] **31** (3.5 g, 28.6 mmol) was distilled (oil bath at 120 °C) into the FVP tube under a stream of N_2 (30 mL/min) with the hot tube heated at 550 °C. After an analogous workup procedure, the crude product was found to crystallize from ether and hexanes. Crystalline lactone **33** [2.3 g, 66%; mp 89–90 °C (lit.[27] mp 89.0–90.2 °C); reported yield, 85%] was obtained in satisfactory yield the first time with no attempts at optimization. This establishes that the low yield of the δ-lactone **27** described in the earlier experiment is probably a characteristic of the molecule rather than the FVP apparatus used in this study.

9,10-Seco-2-oxacholesta-5(10),8,25-trien-6-yn-1-one (26). The lactone **27** (163 mg, 1.2 mmol) was added under nitrogen to a mixture of DMF (1 mL), Et_3N (0.55 mL, 3.9 mmol) and bis-triphenylphosphine-palladium dichloride (18 mg, $Pd(PPh_3)_2Cl_2$, 2 mol %). The solution was heated at 75 °C at which point triflate **10** (394 mg, 1.0 mmol) in 1 mL of DMF (2 × 0.5 mL rinsings) was added dropwise via cannula. Heating at 75 °C was continued for 4.5 h and then the solution was cooled and diluted with ether. The ether layer was separated and then washed successively with 1 M H_2SO_4, saturated $NaHCO_3$, H_2O and brine. Drying over $MgSO_4$ and evaporation of solvent in vacuo gave the desired coupled product, which was purified by flash chromatography

(37) We are grateful to S. Isaeff and G. Syn for performing most of the ozonolyses of vitamin D_2.

(38) Prepared from 3-methyl-3-buten-1-ol by the method of Roberts: Cox, E. F.; Caserio, M. C.; Silver, M. S.; Roberts, J. D. *J. Am. Chem. Soc.* **1961**, *83*, 2719. It is obtained as a clear liquid (bp 103 °C).

(39) The structure was assigned on the basis of the similarity of its ^1H NMR to that of the parent 14-epi-Grundmann's ketone: Condran, P. C., Jr.; Hammond, M. L.; Mouriño, A.; Okamura, W. H. *J. Am. Chem. Soc.* **1980**, *102*, 6259.

(10% EtOAc/hexanes) yielding 309 mg (81%) of **26** as a colorless viscous oil.

(6*Z*)-9,10-Seco-2-oxacholesta-5(10),6,8,25-tetraen-1-one (35). A solution of acetylenic lactone **26** (193 mg, 0.51 mmol) and Lindlar catalyst (260 mg) in 25 mL of hexanes was exposed to hydrogen gas at atmospheric pressure for 1 h. After filtration, the solvent was removed, and a ^{1}H NMR spectrum was obtained. In some cases, the reaction was incomplete and the process was repeated until no starting material remained. In repeating this process, 1-h increments (or less depending on the extent of hydrogenation) of exposure to H_2 gas were used in order to avoid over hydrogenation. Purification by flash chromatography (10% EtOAc/hexanes) followed by HPLC (5% EtOAc/hexanes, 6.0 mL/min) gave the pure tetraene **35** (132 mg, 0.35 mmol) in 68% yield.

2-Nor-1,3-seco-25-dehydro-1-hydroxyvitamin D_3 (37). The previtamin lactone **35** (23 mg, 0.06 mmol) was placed in a flask with 3 mL benzene and cooled to 0 °C. Diisobutylaluminum hydride (0.18 mL, 1.0 M in hexanes) was added dropwise via syringe to the lactone solution with stirring and cooling, and then the solution was allowed to warm to room temperature with stirring over 3 h. The resulting solution was poured onto a mixture of 1.0 M HOAc (0.5 mL) and 10 g of ice. The solution was transferred to a separatory funnel, diluted with ether, and washed with saturated $NaHCO_3$ and brine. Drying over Na_2SO_4, concentration, and flash chromatography (100% EtOAc) afforded 20 mg (83%) of a mixture of the previtamin diol **36** and the vitamin **37**. The pure vitamin **37** is obtained by simply allowing the mixture to stand at ambient temperature for several hours. The ^{1}H NMR exhibits signals that are due only to the [1,7]-sigmatropic shifted vitamin form **37**.

(6*Z*)-9,10-Seco-25-hydroxy-2-oxacholesta-5(10),6,8-trien-1-one (38). The previtamin lactone **35** (7.6 mg, 0.020 mmol) was placed in a flask with THF (0.4 mL) and H_2O (0.1 mL), and then the mixture was cooled to 0 °C. Mercuric acetate (8.8 mg, 0.026 mmol) was added, and the solution was stirred for 4.5 h. A 0.5 M solution of $NaBH_4$ in 1.0 M NaOH was prepared and 50 μL of this reducing solution was added to the reaction mixture. The solution was immediately diluted with ether and transferred to a separatory funnel. The ether layer was washed with saturated $NaHCO_3$ and brine and dried over Na_2SO_4. The crude product was purified by chromatography (35% EtOAc/hexanes), resulting in 5.3 mg (67%) of the 25-hydroxy lactone **38**. Near quantitative recovery of unreacted starting material (~2.3 mg) was also achieved.

2-Nor-1,3-seco-1,25-dihydroxyvitamin D_3 (8). The 25-OH lactone **38** (5.3 mg, 0.013 mmol) was placed in a flask with 0.8 mL of benzene and 0.06 mL of a 1.0 M diisobutylaluminum hydride solution in hexanes. The resulting solution was stirred under argon for 15 h, after which time the solution was diluted with ether and poured onto a mixture of ice (2 mL) and 1 drop of aqueous 1 M HOAc. The mixture was transferred to a separatory funnel and washed with saturated $NaHCO_3$ and brine. Under the reaction conditions the previtamin triol **39** rearranges quantitatively to the vitamin **8** via the usual [1,7]-sigmatropic hydrogen shift. Drying the ether solution over Na_2SO_4 and evaporation of solvent afforded the desired triol, which was purified intially by flash chromatography (100% EtOAc) and subsequently by HPLC (3.0 mL/min, 100% EtOAc) to give 3.5 mg (67%) of pure **8**.

3-Deoxy-2-oxa-25-dehydrovitamin D_3 (40) and 3-Deoxy-2-oxa-25-dehydroprevitamin D_3 (41). To a solution of **37** (11.8 mg, 0.031 mmol) in 0.3 mL of THF at −78 °C was added *n*-BuLi (0.08 mL, 0.13 mmol, 1.60 M in hexanes), and the mixture was stirred for 1 h. The cooling bath was removed and the solution warmed to room temperature over 1 h, at which time *p*-toluenesulfonyl chloride (7.2 mg, 0.039 mmol) was added. After the mixture was stirred for an additional 30 min, the THF was evacuated to dryness under an argon stream, and then 2 mL of DMF was added to the residue. The resulting solution was heated (oil bath) at 60 °C for 12 h. After cooling, the DMF solution was diluted with ether and washed with saturated $NaHCO_3$ and brine. Drying over $MgSO_4$, filtration, and removal of solvent afforded a mixture of ethers **40** and **41** and recovered starting material (^{1}H NMR). The mixture was purified by flash chromatography, first eluting with 5% EtOAc/hexanes to afford 5.2 mg (45%) of the previtamin **41** and vitamin **40** and then eluting with 100% EtOAc to afford 3.1 mg (26%) of the diol **37**. The mixture of previtamin and vitamin could be resolved (partially separated) by HPLC (2% EtOAc/hexanes with two recycles), but complete equilibration between vitamin and previtamin was apparent (^{1}H NMR) in less than 1 h at room temperature. The HPLC–NMR[31] technique was conveniently utilized to identify the first peak as the previtamin **41** and the second as the vitamin **40** as discussed in the text. The experiment was carried out with a Whatman M-9 Partisil column (2% EtOAc/Freon 113 as solvent) on a Waters 6000A HPLC system coupled to a Nicolet 300-MHz NMR spectrometer. The equilibrium ratio was obtained by ^{1}H NMR. The signals due to the angular methyl groups (C_{18}–CH_3) as well as signals due to H_6 in the vitamin and H_9 in the previtamin were integrated. As an additional check of this ratio, the signals due to H_1 were also integrated. The ratio of vitamin to previtamin was determined to be 56/44.

25-Hydroxy-3-deoxy-2-oxavitamin D_3 (5) and 25-Hydroxy-3-deoxy-2-oxaprevitamin D_3 (42). A solution of the equilibrium mixture of previtamin **40** and vitamin **41** (3.8 mg, 0.010 mmol) in a 4:1 mixture of THF/H_2O (0.25 mL) was cooled to 0 °C. Mercuric acetate (4.3 mg, 0.013 mmol) was added, and the solution was allowed to stir at room temperature for 4 h. The reaction was quenched with 25 μL of a 0.5 M solution of $NaBH_4$ prepared in 1.0 M NaOH in methanol. The resulting solution was diluted with ether, washed successively with saturated $NaHCO_3$ and brine, and then dried over $MgSO_4$. After removal of solvent the crude product was purified by flash chromatography eluting with 10% EtOAc/hexanes to afford 0.9 mg (22%; 50% yield based on recovered starting material) of a mixture of hydroxylated vitamin and previtamin ethers **5** and **42**, respectively, and 2.1 mg (57%) of recovered starting material, which could be recycled.

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Supplementary Material Available: Spectral and analytical data for the compounds in the Experimental Section (13 pages). Ordering information is given on any current masthead page.