droxide in 10 mL of DMF produce a pale white suspension which was warmed to 60 °C for 1 h. After the reaction mixture cooled, it was added slowly to 50 mL of ice-water. The solution was washed with CH_2Cl_2 (3 × 20 mL) and acidified by using CH_3CO_2H. The insoluble material was compound **4a** (0.80 g, 95%).

Method B. Similarly, **2a** in ethanol with sodium ethoxide at 50 °C for 1 h, followed by evaporation to dryness and trituration with aqueous CH_3COOH, gave **4a** as a white solid.

3-Nitroimidazo[1,2-*a*]pyridine (7) was prepared according to Paolini.[2d]

2-Methyl-3-nitroimidazo[1,2-*a*]pyridine (8) was prepared according to Hand.[1c]

Attempts to Displace the Nitro Group from 7 and 8. Treatment of 1.63 g (0.01 mol) of **7** with ethyl thioglycolate in 60 mL of DMF and 0.24 g of lithium hydroxide as in the general procedure produced a pale yellow solution which was poured into 200 mL of ice-cold water. The solid was collected and extracted with CH_2Cl_2. Evaporation of the solvent yielded the starting material (1.55 g, 95% recovery by ^1H NMR).

Similarly, the reaction of 1.77 g (0.01 mole) of **8** with $NaSCH_2CO_2Et$ and LiOH does not proceed (1.61 g, 91% recovery by ^1H NMR).

Attempts to Displace the Nitro Group from 9. Treatment of 2.80 g (0.01 mol) of **9** with ethyl thioglycolate in 100 mL of DMF and 0.24 g of lithium hydroxide produced a complex mixture of unidentified products.

3-Amino-2-carbethoxyimidazo[1,2-*a*]pyridine (10). To a stirred solution of **1a** (2.35 g, 0.01 mol) in DMF (20 mL) at −5 °C was added all at once a solution of 0.7 g of purified NaSH in 20 mL of DMF. The dark green solution was allowed to stand to room temperature about 10 min, and the stirring was continued for 30 min. The DMF solution was poured into 300 mL of ice-water. A solid (1.2 g) was collected by filtration and was subjected to chromatography on alumina. Elution with CH_2Cl_2 gave **10** (0.81 g). After the filtrate was evaporated, the resulting product (0.42 g) was treated with CH_2Cl_2 and was sujected to chromatography on alumina to give 0.35 g of additional **10**: 1.16 g total (56.6%); mp 210–212 °C; ^1H NMR ($CDCl_3$, relative to external Me_4Si) δ ~5.2 (NH_2), 2.81 (CH_3), 4.45 (CH_2), 6.71 (H-6), 7.06 (H-7), 7.48 (H-8), 7.76 (H-5).

Anal. Calcd for $C_{10}H_{11}O_2N_3$: C, 58.54; H, 5.36; N, 20.48. Found: C, 58.51; H, 5.37; N, 20.50.

Acknowledgment. We are indebted to Dr. W. W. Paudler and E. H. Hand, Department of Chemistry, University of Alabama, for their helpful discussions in the preparation of the manuscript and thank H. Viols for his technical assistance.

Registry No. 1a, 62223-44-3; **1b**, 67625-26-7; **1c**, 72721-16-5; **1d**, 67625-30-3; **1e**, 67625-29-0; **1f**, 76156-94-0; **1g**, 67625-34-7; **2a**, 76156-95-1; **2b**, 76156-96-2; **2c**, 76156-97-3; **2d**, 76156-98-4; **2e**, 76156-99-5; **3a**, 76157-00-1; **3b**, 76157-01-2; **3c**, 76172-94-6; **3d**, 76157-02-3; **3e**, 76157-03-4; **3f**, 76157-04-5; **4a**, 76157-05-6; **4b**, 76157-06-7; **4c**, 76157-07-8; **4d**, 76157-08-9; **4e**, 76157-09-0; **5a**, 76157-10-3; **5b**, 76157-11-4; **5c**, 76157-12-5; **10**, 76157-13-6; ethyl 7-methylimidazo[1,2-*a*]pyridine-2-carboxylate, 70705-33-8; ethyl thioglycolate, 623-51-8.

A New Synthesis of 7-Dehydrocholesterols

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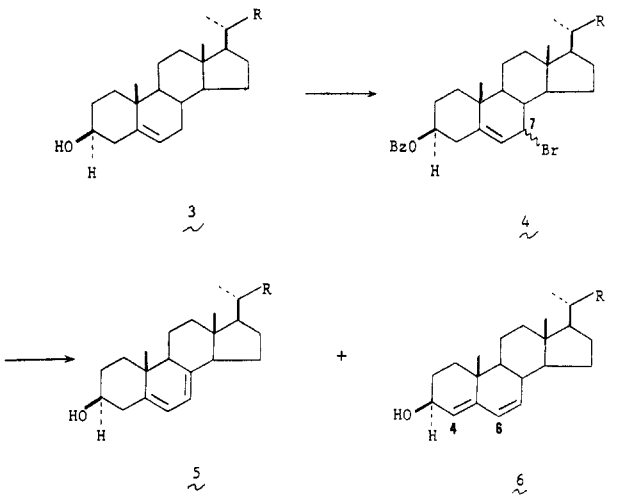
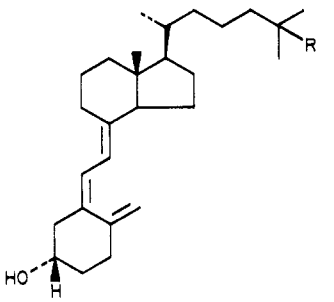
In conjunction with calcitonin and parathyroid hormone, vitamin D and its metabolites are largely responsible for the critical maintenance of calcium homeostasis.[1] The isolation and structural elucidation of a host of biologically active metabolites of vitamin D_3 (cholecalciferol) (1) have been the result of intense investigations begun in the early sixties,[2] highlighted by the landmark discovery of the first human metabolite, 25-hydroxy vitamin D_3 (**2**), by DeLuca.[3] These results soon triggered a corresponding effort to develop both total[4] and partial syntheses of all the known metabolites of vitamin D_3 as well as a number of analogues.[5] To date, all of the preparatively useful approaches to these compounds require the conversion of a cholesterol derivative such as **3** to its 7-dehydro counterpart **5** (Scheme I). Such a transformation was described in 1942 in the classical paper of Ziegler[6] which dealt with allylic bromination. Treatment of a suitable cholesterol ester with NBS afforded a 7-bromo derivative such as **4**, obtained as a mixture of epimers at C(7). The subsequent dehydrobromination and hydrolysis led to 7-dehydrocholesterol **5** along with a substantial quantity of the undesired 4,6-diene isomer **6**. The contaminant **6** has plagued this conversion despite almost 40 years of exhaustive developmental studies.[7] We report a process which affords

Scheme I





1, R = H
2, R = OH

(1) Norman, A. W. "Vitamin D—The Calcium Homeostatic Steroid Hormone"; Academic Press: New York, 1979.

(2) Norman, A. W.; Lund, J.; DeLuca, H. F. *Arch. Biochem. Biophys.* **1964**, *108*, 12. Lund, J.; DeLuca, H. F., *J. Lipid Res.* **1966**, *7*, 739. Haussler, M. R.; Norman, A. W. *Arch. Biochem. Biochem. Biophys.* **1967**, *118*, 145.

(3) Blunt, J. W.; DeLuca, H. F.; Schnoes, H, K. *Biochemistry* **1968**, *7*, 3317. Blunt, J. W.; DeLuca, H. F.; Schnoes, H. K. *Chem. Commun.* **1968**, 801.

(4) Kocienski, P. J.; Lythgoe, B.; Waterhouse, I. *Tetrahedron Lett.* **1979**, 4419–22 and leading references therein.

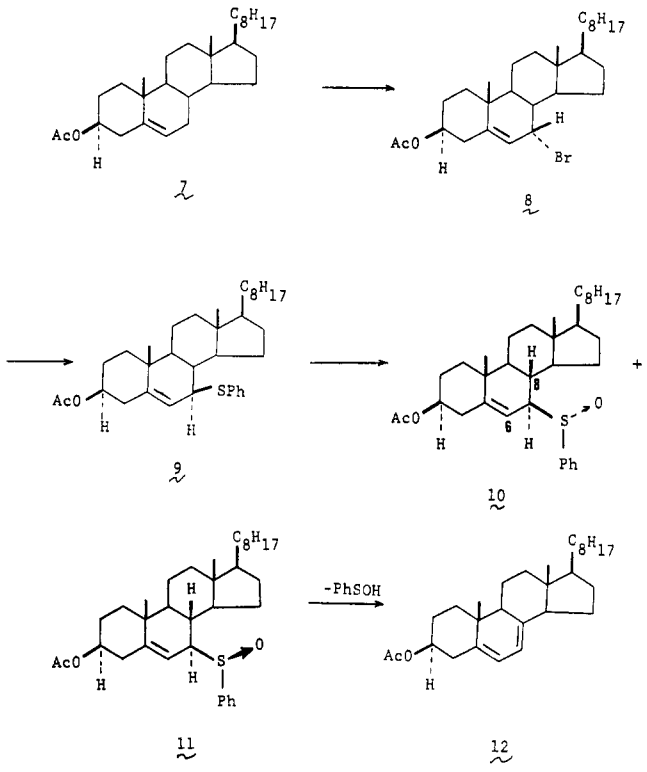
(5) See ref 1 for the various syntheses of the metabolites of vitamin D_3 (pp 95–110) and a number of analogues (pp 111–116).

(6) Ziegler, K.; Späth, A.; Schaaf, E.; Schumann, W.; Winkelmann, E. *Justus Liebigs Ann. Chem.* **1942**, *551*, 80–119.

(7) Tachibana, Y. *Bull. Chem. Soc. Jpn.* **1978**, *51*, 3085–86 and leading references therein. See also: Bernstein, S.; Binovi, L. J.; Odrfman, L.; Sax, K. J.; Subbarow, Y. *J. Org. Chem.* **1949**, *14*, 433–46; Bernstein, S. U.S. Patent 2 498 390, 1950; Pickholz, U.S. Patent 2 568 025, 1951.

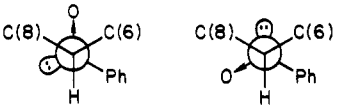
0022-3263/81/1946-1030$01.25/0

Scheme II



pure 7-dehydrocholesterols and avoids entirely the production of any 4,6-diene species.[8]

Treatment of cholesterol acetate (7) with dibromantin/sodium bicarbonate in refluxing hexane yielded the expected mixture of 7-bromides with a 7α/7β ratio[9] of 1.15 (Scheme II). These epimers were then equilibrated in a subsequent step with excess lithium bromide to afford predominantly the desired 7α bromide 8 (7α/7β ratio 4.0).[10] Interestingly, a recent patent[11] claims a similar equilibration of 7-bromocholesterols employing only selected solvents and temperatures. However, we have found that under these patented conditions the 7α/7β bromide ratio remains at 1.15, indicating that free bromide is a requirement for this process.[12] Reaction of the predominant 7α-bromide 8 with benzenethiol yielded the 7β-phenyl sulfide 9 which was oxidized to a readily separable mixture of the (S)-sulfoxide 10 and the (R)-sulfoxide 11 in a ratio of 1:2, respectively. The obtention of the (R)-sulfoxide 11 as the major diastereomer is consistent with expectations based upon kinetic control of the oxidation. Attack of the peroxidizing species presumably occurs from the less hindered side of the most stable sulfide conformation, leading to the predominance of 11. The structures of the sulfoxides were determined by the following NMR analysis. As shown by Nishio,[13] the chemical shift of a proton attached to a carbon α to a sulfoxide is influenced in a predictable fashion by the stereochemistry of the sulfoxide itself. Thus, the C(7) hydrogen of the (R)-sulfoxide 11 was found to absorb at δ 3.59 ($CDCl_3$), whereas the corresponding chemical shift of the C(7) proton of the (S)-sulfoxide 10 occurred at δ 3.01 ($CDCl_3$). This is consistent with a trans relationship of the C(7) hydrogen to the sulfoxide oxygen in the major sulfoxide 11 (Newman projection A) and a gauche relationship in 10 (Newman projection B). These exact orientations are to be found



A B

when the sterically demanding phenyl group resides on the more accommodating α side of the steroidal plane, adopting a preferential trans relationship to C(8). Models readily show this to be the lowest energy conformation of the sulfoxides.

Further support for these assignments was based upon the observation that the C(6) olefinic proton appearing at δ 5.9 in the (R)-sulfoxide 11 is shifted to δ 4.8 in the (S)-sulfoxide 10. This is a consequence of the increased *deshielding* of the C(6) hydrogen by the sulfoxide oxygen in structure 11 vs. 10, a result of the strongly interacting gauche relationship in the former case.

The assumption of kinetic control was readily supported by existence of a facile thermal interconversion of these sulfoxides at 50 °C to an equilibrium mixture of approximately 1:1, starting with either pure compound. Therefore, separation of 10 and 11 was not at all necessary in practice, and at 70 °C a smooth cis elimination of phenylsulfenic acid occurred across C(7)–C(8) to afford the desired 7-dehydrocholesterol ester 12[14,15] The total absence of the 4,6-diene isomer was readily demonstrated by the UV examination of product and mother liquors, showing none of the characteristic absorption of the contaminant.

The fate of the minor amount of 7β-bromide 13, which is also carried through this sequence, is depicted in Scheme III. These results were rigorously demonstrated by working with pure 7α-thiophenoxycholesterol acetate 14, obtained by reaction of the unequilibrated 7-bromides with benzenethiol followed by chromatographic purification. Oxidation of the sulfide 14 with MCPBA yielded the 7α-sulfoxides 15. Lacking the required cis relationship to the C(8) hydrogen, the sulfoxides 15 did not eliminate to 7-dehydrocholesterol acetate, but rather afforded the alcohols 18 upon thermolysis.[16] This reaction, which occurred

(8) For existing methodology designed to accomplish this end, see: Dauben, W. G. *J. Org. Chem.* **1971**, *36*, 3277; Caglioti, L. *Chim. Ind. (Milan)* **1963**, *559*; Salmond, W. G.; Barta, Cain, A. M.; Sobala, M. C. *Tetrahedron Lett.* **1977**, 1683–6; ref 11.

(9) We developed a simple analytical technique to measure the absolute amounts of 7α- and 7β-bromocholesterols. The mixture is treated with benzenethiol/triethylamine (1:1 equiv) in methylene chloride for 1 h at 25 °C. The resulting 7α- and 7β-phenyl sulfides are then quantitatively separated by chromatography over silica, eluting with methylene chloride/hexane, 1:1. The validity of this technique was shown by the *quantitative* conversion of pure 7α-bromocholesterol benzoate to its 7β-phenyl sulfide derivative with *100% inversion*, thus indicating that the relative amounts of derived sulfides relate directly to the quantities of bromo precursors.

(10) Similarly, treatment of the bromide mixture with sodium iodide yielded the 7-iodo derivative with a 7α/7β ratio of 4.0 also. No advantage over the bromide series was therefore obtained by working with the more sterically demanding C(7) iodides.

(11) Salmond, W. G. U.S. Patent 3970676, July 20, 1976.

(12) Treatment of pure crystalline 7α-bromocholesterol benzoate with excess sodium iodide yielded 7α-iodocholesterol benzoate, mp 115 °C dec (benzene/acetone), as the major (80%) product. The observed retention of configuration is clearly a result of the postulated halide-catalyzed equilibration.

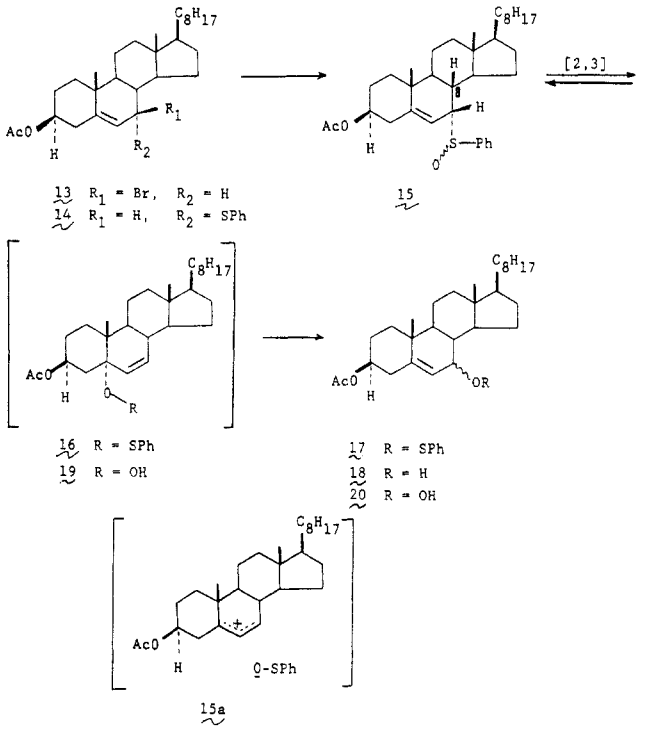
(13) Nishio, M. *Chem. Commun.* **1969**, 52–52.

(14) The product was identical in all respects with authentic sample prepared according to the method described: Norman, A. W., Schaefer, K., Coburn, J. W., DeLuca, M. F., Fraser, D., Grigoleit, H. G., Herrath, D. V., Eds. "Vitamin D—Biochemical, Chemical, and Clinical Aspects Related to Calcium Metabolism"; Walter de Gruyter: New York, 1977; pp 51–54.

(15) Interestingly, the corresponding sulfone, prepared from the sulfide 9 by oxidation with 2 equiv of MCPBA, was thermally stable to at least 200 °C.

(16) The structural assignments of the alcohol 18 follow readily from spectral data, particularly NMR ($CDCl_3$) which exhibits only *one* olefinic proton at δ 5.7–5.4, with the C(7) proton appearing at δ 3.85, thereby ruling out the 5-hydroxy-Δ^6 isomeric series. See: Henbest, H. B.; Jones, E. R. H. *J. Chem. Soc.* **1948**, 1792.

Scheme III



smoothly at 100 °C in toluene (vs. only 70 °C for the conversion of **10, 11 → 12**), presumably proceeds via the ion pair **15a**. This transient intermediate may result from dissociation of either the 7α-sulfoxide **15** or the Δ⁶-5α-phenylsulfenate ester **16**, expected to be in thermal equilibrium with **15**, interconverting by a facile [2,3] sigmatropic rearrangement. Eventual collapse of the ion pair **15a** could then generate the Δ⁵-7α,β-sulfenates **17**, yielding the observed alcohols **18** upon hydrolytic workup.[17] This pathway is precedented in the singlet-oxygen reaction[18] of cholesterol which yields the isolable Δ⁶-5α-hydroperoxide **19**, an unstable species which slowly rearranges on standing to the epimeric mixture of 7α,β-hydroperoxides **20**.

In practice, the overall conversion of cholesterol esters to their 7-dehydrocholesterol derivatives proceeds smoothly without the need to purify any of the intermediates. The desired 7-dehydrocholesterol ester (originating ultimately from the predominant 7α-bromide of type 8) is separated from the minor byproduct alcohols, e.g., **18** (derived from the 7β-bromide species **13**), by a simple filtration over silica. The overall yields are on average over 50%. This methodology is quite general and has been applied to a variety of cholesterol derivatives relevant to the synthesis of the human metabolites of vitamin D_3. We therefore believe that this process for the synthesis of 7-dehydrocholesterols offers a viable alternative to the existing technology for the preparation of these extremely important substances.

Experimental Section

Melting points were determined on a Rinco Model M-50 melting-point apparatus and are uncorrected. IR spectra were obtained by using a Beckmanan IR-9 spectrophotometer. A Cary 14 recording spectrophotometer was used for UV absorption spectra. NMR spectra were determined with Varian T-60 and HA-100 spectrometers, using tetramethylsilane as the internal reference. Mass spectra were recorded on a CEC 21-110B mass spectrometer at 70 eV, using a direct insertion probe. Thin-layer chromatography was carried out by using Merck F-254 silica gel plates. The general overall procedure will be exemplified by the synthesis of 7-dehydrocholesterol acetate (**12**) from cholesterol acetate (**7**).

7-Dehydrocholesterol Acetate (12). A mixture of 50 g (0.117 mol) of cholesterol acetate (**7**), 23.82 g (0.082 mol) of dibromantin, and 53.04 g (0.631 mol) of sodium bicarbonate in 2 L of hexane was heated under reflux (argon) for 0.5 h. The reaction was cooled and filtered to remove 5,5-dimethylhydantoin (and inorganic salts), and the filtrate was evaporated to dryness. The residue was taken up in 400 mL of toluene and treated with 20.32 g (0.233 mol) of anhydrous lithium bromide in 270 mL of acetone. The mixture was stirred at 0 °C for 2 h, removed from the ice bath, and treated with 22.1 mL (0.157 mol) of triethylamine and 16.0 mL (0.157 mol) of benzenethiol. After being stirred for 1.25 h at 25 °C, the reaction was diluted with 1 L of ethyl acetate, washed with 500 mL of 1 N HCl, and two 500-mL portions of water. The organic phase was dried over sodium sulfate and evaporated.[19] The residue was dissolved in 770 mL of ethyl acetate, cooled to 0 °C, and treated with 26.05 g (0.128 mol) of *m*-chloroperbenzoic acid (85%) for 2 h. The mixture was washed with 10% $NaHCO_3$ and water. The organic phase was dried over sodium sulfate and evaporated.[20] The residue was dissolved in 1 L of toluene, treated with 36 mL (0.256 mol) of triethylamine, heated at 70 °C for 28 h, cooled, and washed twice with water. The organic phase was dried over sodium sulfate and evaporated. The residue was filtered through 1.5 kg of silica, eluting with methylene chloride. fractions containing the product were combined and evaporated to yield 33.97 g (68%) of 7-dehydrocholesterol acetate (**12**). The product was recrystallized from methylene chloride/methanol to afford 26.56 g (53%) of pure **12**: white needles; mp 129–130 °C (lit.[21] mp 129–130 °C); IR (KBr) 2950, 1735 (OAc), 1260 cm^{-1}; NMR ($CDCl_3$) δ 5.56, 5.48 [br q, 2 H, C(6)-H, C(8)-H], 4.7 (br m, 1 H, CHOAc), 2.03 (s, 3 H, Ac); mass spectrum *m/e* 426 (M^+),366 (M^+ – HOAc), 351, 281, 253; UV max (hexane) 271 nm (ε 11 620), 281 (12 320), 294 (7050). Anal. Calcd for $C_{29}H_{46}O_2$ (mol wt 426.694): C, 81.63; H, 10.87. Found: C, 81.58; H, 10.78.

Acknowledgment. We thank the staff of the Physical Chemistry Department of Hoffmann-La Roche Inc. for the determination of spectral and analytical data.

Registry No. 7, 604-35-3; 12, 1059-86-5.

(17) Esters of sulfenic acids (RSOH) are known to be very readily hydrolyzed. See: Kharasch, N.; Potempa, S. J.; Wehrmeister, H. L.; *Chem. Rev.* **1946**, *39*, 323–26.

(18) Schenck, G. O.; Neumueller, O. A.; Eisfeld, W. *Justus Liebigs Ann. Chem.* **1958**, *618*, 202–210.

(19) The pure 7β-phenyl sulfide **9** may be obtained at this point as a white solid, mp 99–100 °C (2-propanol), by chromatography over silica, eluting with methylene chloride/hexane, 1:1.

(20) The sulfoxides **10** and **11** may be isolated at this point by chromatography over silica, eluting with methylene chloride/ethyl acetate, 9:1. The (*R*)-sulfoxide **11** was obtained as white needles, mp 128–130 °C (CH_3OH); the (*S*)-sulfoxide **10** was an amorphous white solid.

(21) Schaltegger, H. *Helv. Chim. Acta* **1950**, *33*, 2101–2110.

Diastereoisomers of 3-Methylpyroglutamic Acid and β-Methylglutamic Acid

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Compared with numerous publications concerning α- and γ-methylglutamic acids, reference to β-methylglutamic acid in the literature is relatively scant. Although several syntheses have been reported [1-6], none describes the sep-

(1) J. Smrt and F. Šorm, *Collect. Czech. Chem. Commun.*, **18**, 131 (1953).

(2) A. Meister, L. Levintow, R. E. Greenfield, and P. A. Abendschein, *J. Biol. Chem.*, **215**, 441 (1955).

