absorption measurements, the concentration was adjusted so that ~50% of the light was absorbed. Lower concentrations were used for emission studies to avoid any problems of reabsorption.

Pyrex NMR tubes (5 mm) were used to hold the samples for the emission experiments. Rectangular (2 × 10 mm) Pyrex cells were used for the absorption measurements. All samples were immersed directly into liquid helium in a Pyrex double Dewar, with windows in the liquid-nitrogen jacket to avoid passing the light through boiling nitrogen.

All absorption and emission spectra were obtained on a 1-m Jarrel-Ash spectrometer. Site selection spectra of azulene and chrysene were obtained using the 3511-Å line of the argon-ion laser. Absorption spectra were obtained with a quartz-halogen lamp. The excitation source for the broad-band emission spectra was a high-pressure mercury lamp in combination with broad-band filters. Site selection spectra were obtained with a tunable continuous-wave jet-flow dye laser pumped with an 8-W argon-ion laser. Rhodamine 110, pumped with all visible lines, could be tuned from ~530 to 595 nm. Coumarin 1, pumped with the UV lines, was tunable from 455 to 485 nm. Intracavity etalons were used to reduce the output bandwidth to less than 1 cm^{-1}.

The curves displayed in Figures 3–5 were generated and plotted on an IBM-1130 computer.

References and Notes

(1) This paper is based primarily on the Ph.D. Thesis of William C. McColgin, University of Rochester, 1975.
(2) (a) Eastman Kodak Co.; (b) JILA Visiting Fellow, 1977–1978; (c) Joint Institute for Laboratory Astrophysics.
(3) J. H. Eberly, W .C. McColgin, K. Kawaoka, and A. P. Marchetti, *Nature (London)*, **251**, 215 (1974).
(4) W. C. McColgin, Ph.D. Dissertation, Department of Physics and Astronomy, University of Rochester, 1975.
(5) R. I. Personov, E. I. Al'shitz, and L. A. Bykovskaya, *Zh. Eksp. Teor. Fiz., Pis'ma Red.*, **10**, 609 (1972); *Opt. Commun.*, **6**, 169 (1972).
(6) R. I. Personov, E. I. Al'shitz, L. A. Bykovskaya, and B. M. Kharlamov, *Zh. Eksp. Teor. Fiz.*, **65**, 1825 (1973); *Sov. Phys.-JETP (Engl. Transl.)*, **38**, 912 (1974).
(7) K. Cunningham, J. M. Morris, J. Funfschilling, and D. F. Williams, *Chem. Phys. Lett.*, **32**, 581 (1975).
(8) I. I. Abram, R. A. Auerbach, R. R. Birge, B. E. Kohler, and J. M. Stevenson, *J. Chem. Phys.*, **63**, 2473 (1975).
(9) E. I. Al'schitz, R. I. Personov, and B. M. Kharlamov, *Chem. Phys. Lett.*, **40**, 166 (1976), and references cited therein.
(10) R. I. Personov and B. M. Kharlamov, *Opt. Commun.*, **7**, 417 (1973).
(11) B. M. Kharlamov, R. I. Personov, and L. A. Bykovskaya, *Opt. Spectrosc.*, **39**, 137 (1976); *Opt. Commun.*, **12**, 191 (1974).
(12) A. A. Gorokhovski and L. A. Rebane, *Opt. Commun.*, **20**, 144 (1977).
(13) A. P. Marchetti, M. Scozzafava, and R. H. Young, *Chem. Phys. Lett.*, **51**, 424 (1977).
(14) The apparent growth of the phonon-like sideband is not due to differences in the sites, but reflects the increasing difficulty of selecting only a single site when exciting on the high-energy side of a given absorption transition-where a greater percentage of the absorption results from the broad overlapping phonon sidebands. See ref 4 and 8.
(15) E. V. Shpol'skii, *Sov. Phys. Usp.*, **5**, 522 (1962); **6**, 411 (1963).
(16) E. V. Shpol'skii, *Pure Appl. Chem.*, **37**, 183 (1974), and references cited therein.
(17) G. J. Small, *J. Chem. Phys.*, **52**, 656 (1970).
(18) K. K. Rebane, "Impurity Spectra of Solids", Plenum Press, New York, N.Y., 1970.
(19) A. Maradudin, "Solid State Physics", Supplement 3, 2nd ed, F. Seitz and D. Turnbull, Ed., Academic Press, New York, N.Y., 1971, Chapter 8.
(20) A. Szabo, *Phys. Rev. Lett.*, **25**, 925 (1970); **27**, 323 (1971).
(21) L. A. Riseberg, *Phys. Rev., Sect. A*, **7**, 671 (1974).
(22) D. B. Fitchen in "Physics of Color Centers", W. B. Fowler, Ed., Academic Press, New York, N.Y., 1968, Chapter 5.
(23) B. Meyer, "Low Temperature Spectroscopy", American Elsevier, New York, N.Y., 1971, Chapters 1 and 2.
(24) R. M. Hochstrasser and P. N. Prasad in "Excited States", Vol. 1, E. C. Lim, Ed., Academic Press, New York, N.Y., 1974, pp 79–126.
(25) One of the justifications for this broad-band synthesis lies in the fact that the same site model (starting with eq 1) successfully predicts the observed features and behavior of narrow site selection spectra as has been experimentally verified using a tunable dye laser. See ref 4 and 8.
(26) Reference 18, pp 39–49.
(27) I. Abram, R. A. Auerbach, R. R. Birge, B. E. Kohler, and J. M. Stevenson, *J. Chem. Phys.*, **61**, 3857 (1974).
(28) A. P. Marchetti, W. C. McColgin, and J. H. Eberly, *Phys. Rev. Lett.*, **35**, 387 (1975).
(29) The minimum bandwidth in organic solutions appears to be about 0.6 kcal or 250 cm^{-1} fwhm.
(30) G. G. Guilbault, "Practical Fluorescence, Theory, Methods, and Techniques", Marcel Dekker, New York, N.Y., 1973, p 9.
(31) Reference 22, p 309.
(32) The large Stokes shift observed for R6G and resorufin presumes in our model larger values of Δ—on the order of 200 cm^{-1} for resorufin and 300 cm^{-1} for R6G.
(33) G. Flatscher, K. Fritz, and J. Friedrich, *Z. Naturforsch. A*, **31**, 1220 (1976).
(34) Reference 18, pp 60–61.

Conformational Equilibria in Vitamin D. Synthesis and ^1H and ^{13}C Dynamic Nuclear Magnetic Resonance Study of 4,4-Dimethylvitamin D_3, 4,4-Dimethyl-1α-hydroxyvitamin D_3, and 4,4-Dimethyl-1α-hydroxyepivitamin D_3

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Abstract: The title compounds were synthesized and their dynamic properties investigated by variable temperature ^1H NMR. Each of these compounds gave separate signals, at low temperature, for ring A conformers. The temperature-dependent spectra allowed the determination of the activation parameters characteristic of ring A chair–chair interconversion. The free energies of activation ($\Delta G^{\ddagger}$) for the chair inversion in 4,4-dimethylvitamin D_3 (**4a**), 4,4-dimethyl-1α-hydroxyvitamin D_3 (**5**), and 4,4-dimethyl-1α-hydroxyepivitamin D_3 (**6**) were found to be 10.1, 11.0, and 12.0 kcal/mol, respectively. The ^{13}C NMR spectrum of **4a** was recorded at room temperature and at low temperature (ca. −90 °C). The chemical shift separation of the two observed C_3 signals, at low temperature, was used for conformational analysis.

Introduction

Vitamin D is a steroid, whose actual shape considerably differs from other steroids. The difference lies in its cleaved ring B and its C_6–C_7 single bond having an s-trans instead of an s-cis conformation. It appears that this extended structure of vitamin D has the lowest ground state free energy; however, this bond is free to rotate, enabling vitamin D to achieve the

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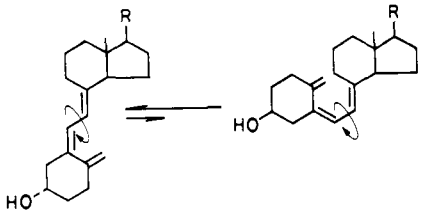


Figure 1. Conformational equilibria: rotation around C_6–C_7 single bond in vitamin D_3 (**1a**) and previtamin D_3.

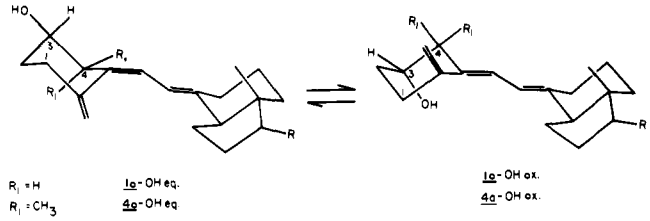
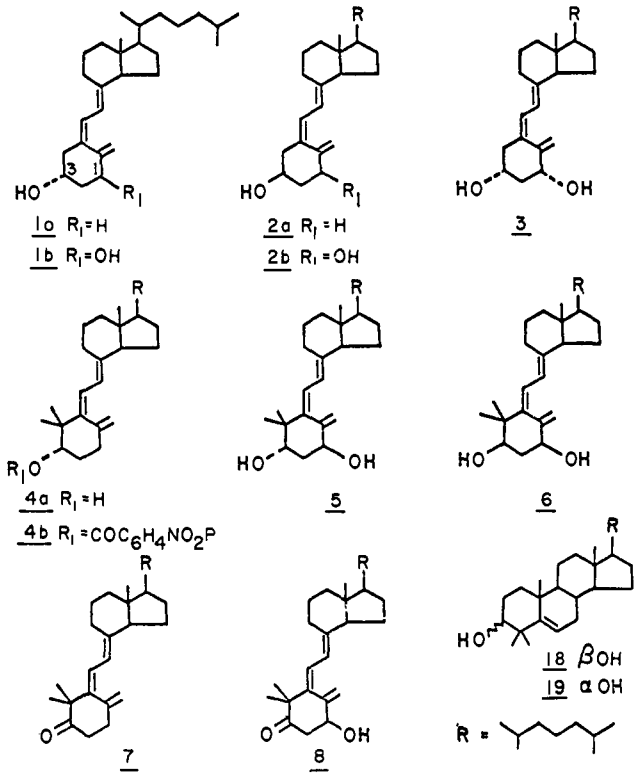


Figure 2. Conformational equilibria: ring A chair–chair interconversion in vitamin D_3 (**1a**) and 4,4-dimethylvitamin D_3 (**4a**).

alternative s-cis conformation required for the vitamin D ⇄ previtamin D thermal equilibrium (Figure 1).[1] This equilibrium is essential for the biogenetic formation of vitamin D in most organisms. The other conformational mobility taking place in vitamin D and its analogues is the widely discussed ring A chair–chair interconversion where two conformers of similar free energy, one with OH at C_3 in an axial and the other in an equatorial orientation, exist in a dynamic equilibrium (Figure 2).[2-5]

An analysis of the ring A conformational equilibrium was performed by ^1H NMR spectroscopy,[2,3] using the values of the spin–spin coupling constants of the proton at C_3 (the ring A carbon bearing an OH substituent). Recently, we have also applied ^{13}C NMR, using a method based on the chemical shift values of the C_3 carbon atom.[5]

It was established that at room temperature, the relative population of ring A conformations in the monools, vitamin D_3 (**1a**) and its C_3 epimer **2a**, as well as in the 1,3-trans diol,



1α-OH vitamin D_3 (**1b**), does not differ appreciably, the ratios of the conformers (OH-equatorial/OH-axial at C_3) being 57:43, 65:35, and 45:55, respectively.[3,5] On the other hand, in nonpolar solvents, the two 1,3-cis diols, 1α-OH epivitamin D_3 (**2b**) and 1β-OH vitamin D_3 (**3**), assume mainly the conformation in which both OH groups are axially oriented, the ratio of the C_3 OH-equatorial to the OH-axial conformations being 20:80 and 10:90, respectively.[3,6,7]

Ring A conformational mobility of vitamin D_3 (**1a**) and its 1α-OH analogue **1b** is of interest in connection with the biological activity of these compounds. It was proposed by Okamura and Norman[3,8] that in the hormonally active 1α,25-dihydroxyvitamin D_3 (**1b**-25-OH), only the conformation having OH at C_1 in an equatorial orientation has the proper geometry for binding to the protein receptor, a step which is necessary to induce the biological events leading to the calcium transport and mobilization in the body.

Our investigations of the dynamic properties of ring A conformations in vitamin D_3 (**1a**) and its hydroxylated analogue using NMR techniques were hampered by their considerable ring A flexibility. We have been unable to detect any spectral changes either in vitamin D_3 (**1a**) or in 1α-OH vitamin D_3 (**1b**) at low temperatures, down to ca. −100 °C. Therefore, we turned our attention to the 4,4-dimethylvitamin D_3 (**4a**), its *p*-nitrobenzoate ester (**4b**), and the respective diols, the 1α-OH-4,4-dimethylvitamin D_3 (**5**) and its C_3 epimer **6**. We have expected that the additional interactions introduced to the molecule due to the methyl groups at C_4 might increase, considerably, the energy barrier to the ring A inversion, enabling the NMR detection of the separate conformers.

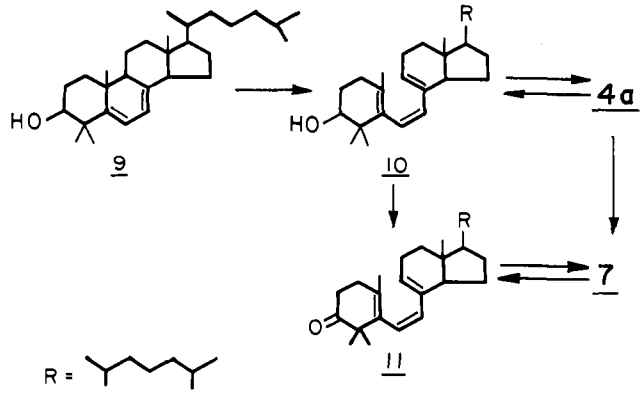
These compounds were thus synthesized and their dynamic properties investigated by variable temperature NMR. In addition, for comparison, we have also prepared the two corresponding dimethyl keto analogues **7** and **8**, and recorded their NMR spectra.

Results

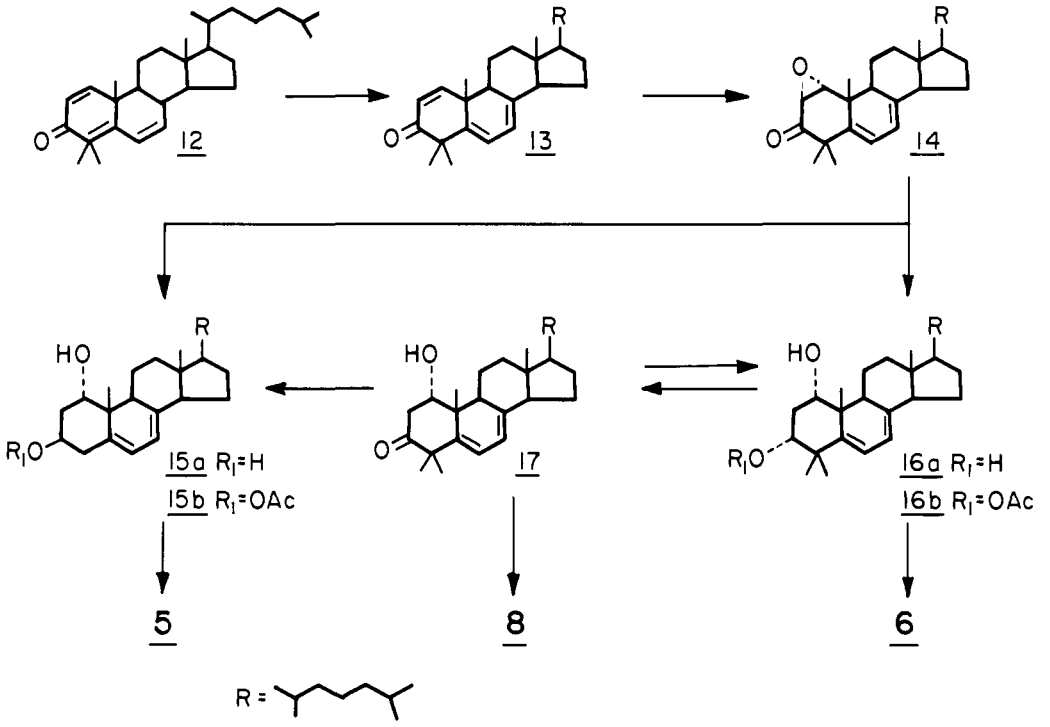
1. Preparation of Compounds. a. 4,4-Dimethylvitamin D_3 (4a), the *p*-Nitrobenzoate Ester (4b), and the Dimethyl Ketone 7. The previously described dimethyl derivative **4a** was prepared by a slight modification of the published procedure (Scheme I).[9] 4,4-Dimethylcholesta-5,7-dien-3β-ol (**9**) was irradiated at 0 °C with a 300-nm light and the previtamin **10** formed was isolated and identified. Heating **10** at 80 °C for 4 h resulted in an equilibrium mixture of previtamin **10** and vitamin **4a** in a 1:9 ratio (as established by ^1H NMR of the heated mixture). The mass spectrum of **4a**[10] shows peaks due to fragmentation across the C_7–C_8 double bond, a unique characteristic of the vitamin D system. However, **4a** absorbs in the UV at a slightly lower wavelength than the parent vitamin D_3 (**1a**), with decreased value (λ_{max} 261 nm, ε 12 000, vs. 264 nm, ε 18 000).

The 4,4-dimethylvitamin D_3 (**4a**) was converted by a stan-

Scheme I



Scheme II



dard procedure to its *p*-nitrobenzoate ester, **4b**. The ester was characterized by its mass spectral fragmentation with peaks due to a loss of *p*-nitrobenzoic acid and a cleavage across the C_7–C_8 double bond,[10] and by its UV spectrum showing a broad band at 256 nm (ϵ 22 000).

Oxidation of **4a** with a modified Moffat reagent[11] led to the keto analogue **7**. This ketone was also formed by similar oxidation of the previtamin **10** followed by thermal isomerization of the resulting keto previtamin analogue **11**. The presence of the triene system in **7** was indicated by its mass spectrum (cleavage across C_7–C_8 bond).[10]

The UV spectrum of **7** showed a number of bands (λ_{max} 320 s, 310 s, 290 s, 264, and 253 nm (ϵ 1200, 2700, 4000, 12 000, and 10 800)) due to the conjugated triene system and the homoconjugation of the carbonyl function with the double bonds.

b. 1α-Hydroxy-4,4-dimethylvitamin D_3 (5), Its C_3 Epimer 6, and Their Keto Analogue 8. These three vitamin D_3 analogues were obtained by irradiation of the corresponding $\Delta^{5,7}$-dienes **15a, 16a,** and **17** formed in three steps from the known trienone **12** as outlined in Scheme II.[12]

The starting material **12**[13] was converted with $NaOCH_3$ and $(CH_3)_2SO$ to its sodium tetraenolate, which gave with methyl iodide the 4,4-dimethyl trienone **13** (λ_{max} 275 nm, ϵ 6000). On epoxidation with methanolic solution of NaOH and H_2O_2 the epoxy ketone **14** (λ_{max} 293 s, 282, 272, 261 s nm (ϵ 5500, 9300, 9300, 7000)) was formed.

The epoxy ketone **14** was reduced with $LiAlH_4$ in boiling ether to a 1:4 mixture of 1α,3β- and 1α,3α-diols **15a** and **16a**.[14] The diol **15a** (multiplet due to H at C_1 and C_3 which separated upon addition of a shift reagent into a narrow, $W_{1/2}$ = 7.5 Hz, and a broader, $W_{1/2}$ = 17 Hz, signal) reacted with acetic anhydride and pyridine at room temperature to give the 3-monoacetate **15b** (H at C_1, a narrow multiplet, $W_{1/2}$ = 7 Hz, and H at C_3, a triplet, J = 8 Hz). On the other hand, the diol **16a** (H at C_1 and C_3, narrow multiplets, $W_{1/2}$ = 7.5 Hz) was not acetylated under the above mild conditions, but gave the 3-monoacetate **16b** (H at C_1 and C_3, two narrow multiplets, $W_{1/2}$ = 7.5 and 7.3 Hz, respectively) with acetic anhydride and *N,N'*-dimethylaminopyridine.

Selective oxidation of the 1α,3α-diol **16a** with a modified Moffat reagent[11] resulted in the ketol **17,** which on treatment with $LiAlH_4$ was reduced to a 1:1 mixture of the two diols **15a** and **16a**.

Both diols **15a** and **16a** as well as the ketol **17** were converted to the respective vitamin D_3 analogues **5, 6,** and **8** by irradiation in ether at 300 nm, followed by heating for 4 h at 80 °C. The two diols **5** and **6** absorb in the UV (λ_{max} 257 nm, ϵ 11 000) at a lower wavelength than their monohydroxylated analogue **4a**. The UV spectrum of the ketol **8** was similar to that of the ketone **7**. In the mass spectra, the three vitamin D_3 analogues showed characteristic peaks due to the fragmentation across the C_7–C_8 double bond.[10]

2. ^{1}H and ^{13}C NMR Spectra. We have investigated the variable temperature ^{1}H NMR spectra of four 4,4-dimethyl derivatives: the monool **4a,** its *p*-nitrobenzoate ester **4b,** and the two 1,3-diols **5** and **6**. We have observed the slow exchange limit spectra for each of the compounds, and determined the activation parameters for their ring A chair–chair interconversion. On the other hand, the two ketones **7** and **8** did not show any spectral changes in the temperature range between 25 and −100 °C.

The ^{13}C NMR spectra of the dimethylvitamin **4a** and its ester **4b** were analyzed, and all their ^{13}C signals were assigned. The low-temperature ^{13}C spectrum of **4a** was also recorded, and the slow exchange limit for the C_3 resonance was observed at ca. −90 °C. The chemical shift difference between the two C_3 signals (due to the two ring A conformations) was compared with the chemical shift difference between the C_3 signals of 4,4'-dimethylcholesterol (**18**) and its C_3 epimer **19**. Since ring A of the two steroids has similar substitution patterns as ring A of **4a** (where **18** has an equatorially oriented OH group and **19** an axially oriented OH), they may be regarded as corresponding to its "frozen" conformers.

The population ratios of the 4,4-dimethyl derivatives were evaluated at room temperature from the coupling constants values of proton at C_3,[2,3] and at the slow exchange limit from the ratio of peak areas. These population ratios were ca. 50:50, remaining constant at low temperatures (within an experimental error of ±10%).

a. ^{1}H NMR of 4,4-Dimethylvitamin D_3 (4a) and Its Ester 4b. The chemical shift data for the vinylic and carbinol protons

Table I. ^{1}H NMR Data for 4,4-Dimethylvitamin-D_3 and Derivatives at Room Temperature[a]

compd	temp, ±2 K	H at C_1, ppm	^3J_c, Hz	^3J_t, Hz	H at C_3, ppm	^3J_c, Hz	^3J_t, Hz	H_E at C_{19}, ppm	J, Hz	H_Z at C_{19}, ppm	J Hz	H at C_7, ppm	H at C_6, ppm	J_{AB}, Hz	$\Delta\nu_{AB}$, ±1.2 Hz
4a	301				3.584	3.7	7.3	4.622	2.7	5.013	1.4	5.92	6.14	11.0	62
4b[b]	299				4.939	3.8	7.2	4.752	2.7	5.127	1.4	6.01	6.25	11.0	64.5
5	287	3.567	3.8*	5.8*	4.207	4.2	8.0	4.789	1.2, 1.6	5.267	2.3, 1.6	5.90	6.26	10.6	96
6	299	3.351	4.5*	4.5*	4.147	4.5*	4.5*	4.793	2.4	5.222	2.5	5.89	6.30	11.1	111

[a] All spectra were recorded on a Bruker WH-270, in CS_2 solution containing CD_2Cl_2 as a lock; chemical shifts are given in parts per million downfield from Me_4Si and are accurate to within ±0.0025 ppm; coupling constants are accurate to within 0.2 Hz; an asterisk indicates coupling constants values obtained by a computer Lorentzian curve fitting. [b] In addition the *p*-nitrobenzoate group shows an A_2B_2 spin system with δ_A 8.13 and δ_B 8.21 ppm, J_{AB} = 8.8 ± 0.2 Hz, and ν_{AB} = 21.5 ± 1.2 Hz.

Table II. ^{1}H NMR Data for 4,4-Dimethylvitamin D_3 and Derivatives at the Slow Exchange Limits[a]

compd	temp, ±2 K	orientation of OH at C_3	H at C_1	H at C_3	H_E at C_{19}	H_Z at C_{19}	H at C_7	H at C_6	J_{AB}, ±0.3 Hz	ν_{AB}, ±2 Hz
4a	181	ax		3.40	4.58	4.99	5.90	6.03	11.3*	36*
		eq		3.21	4.54	4.96	5.77	6.08	10.7*	83*
4b	183	ax		4.99	4.75	5.14	6.00	6.17	11.0*	46*
		eq		4.81[b]	4.69	5.11	5.87	6.25	10.9*	101*
5	208	ax	3.37	4.20	*c*	*c*	*c*	*c*	*c*	*c*
		eq	3.58	4.10	*c*	*c*	*c*	*c*	*c*	*c*
6	226.5	ax		3.88	*c*	5.20	*c*	*c*	*c*	*c*
		eq		4.18		5.16	*c*	*c*	*c*	*c*

[a] See footnote *a*, Table I; chemical shift values are accurate to within ±0.01 ppm. [b] $^3J_{eq:eq}$ = 4.3* ± 0.4 Hz. $^3J_{ax:ax}$ = 11.4* ± 0.4 Hz. [c] These signals were not observed under the slow exchange limit condition.

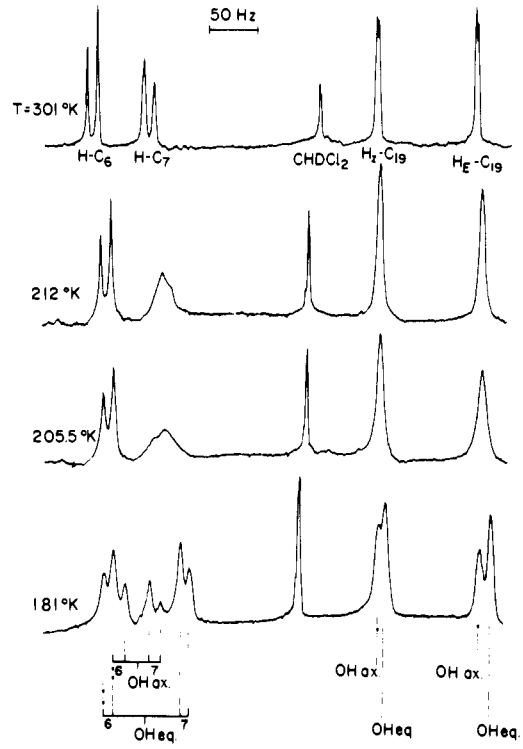


Figure 3. The downfield portion of the 270-MHz spectra of **4a** at several temperatures. The relative shift displacements are due to temperature/solvent shift.

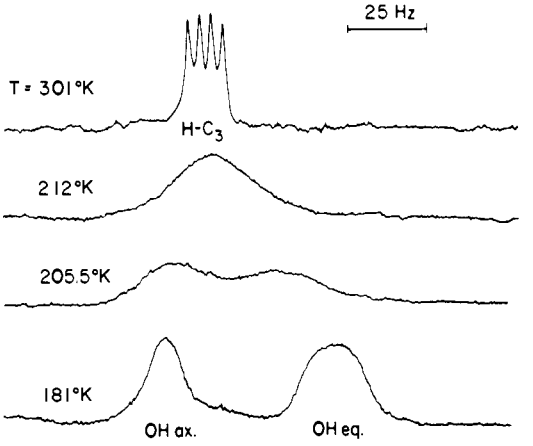


Figure 4. The part of the 270-MHz spectra of **4a** at several temperatures showing the carbinol proton at C_3.

in **4a** and **4b** at room temperature and low temperature are collected in Tables I and II, respectively.

The room temperature spectrum of **4a** shows five downfield multiplets due to H at C_3 (d of d), H_E at C_{19} (d), H_z at C_{19} (d of t), H at C_7 (d), and C_6 (d). All hydrogen multiplets may be considered as a first-order spin system, except for the last two hydrogens, which are part of an AB spin system. The chemical shift differences between the above hydrogens and the neighboring hydrogen atoms, to which they are coupled, satisfy the condition $\delta_A - \delta_B > 3J_{AB}$. As a result, the analysis of these hydrogen multiplets is not complicated by virtual couplings. Thus, the temperature-dependent behavior (Figures 3 and 4), as well as the position of coalescence, can be determined accurately. In the case of the *p*-nitrobenzoate ester, **4b,** we have also monitored the temperature-dependent behavior of the benzoate group's aromatic protons. The true description of the aromatic multiplets at room temperature is an AA'BB' spin system, but as first approximation it can be considered as A_2B_2 spin system with J_{AB} = 8.8 Hz and $\Delta\nu_{AB}$ = 21.5 Hz.

In order to evaluate the activation parameters for the chair-chair interconversion in these compounds, we have used the approximate relation at the coalescence temperature, $k_c = \pi\Delta\nu/\sqrt{2}$, together with the Eyring equation. Upon substitution, rearrangement, and evaluation of the constants involved, the following approximate equation for the free energy

Table III. Thermodynamic Data and Parameters for 4,4-Dimethylvitamin D_3 (**4a**) and Its *p*-Nitrobenzoate Ester (**4b**)

compd	signal obsd for protons at	T_c, ±2 K	k_c at T_c,[a] s^{-1}	$\Delta G_c^{\ddagger}$ at T_c,[b] kcal/mol	$\Delta G^{\ddagger}$,[c] kcal/mol	E_a, kcal/mol	$\Delta H^{\ddagger}$, kcal/mol	$\Delta S^{\ddagger}$ [d]
4a	C_3	208	120	10.06	10.07 ± 0.2	11.14 ± 0.8	10.74 ± 0.8	3.4 ± 3
	C_7	204	73	10.06				
	C_6	197	31	10.03				
	$C_{19\,(E)}$	197	27	10.09	(10.08 ± 0.04)			
	$C_{19\,(Z)}$	195	18	10.14				
4b	C_3	207	104	10.06	10.09 ± 0.2	10.97 ± 0.7	10.57 ± 0.7	2.37 ± 3
	C_7	204	73	10.04				
	meta[e]	202	47	10.13				
	C_6	202	46	10.13				
	$C_{19\,(E)}$	200	38	10.11	(10.10 ± 0.03)			
	ortho[e]	198	29	10.11				
	$C_{19\,(Z)}$	195	20	10.10				

[a] The k_c's were calculated from the equation $k = 2^{-1/2}\pi\Delta\nu$. [b] The $\Delta G_c^{\ddagger}$'s were obtained by use of the equation $\Delta G_c^{\ddagger} = T_c(45.67 + 4.58 \log (T_c/\Delta\nu))$. [c] The $\Delta G_c^{\ddagger}$'s were obtained from a plot of ln k_c vs. $1/T_c$ for all protons involved using the equation $k_c = A \exp(-E_a/RT_c)$; values in parentheses indicate the average $\Delta G_c^{\ddagger}$ calculated from the previous column together with the standard deviation. [d] The $\Delta S^{\ddagger}$'s were obtained from the equation $\Delta S^{\ddagger} = R(\ln(hA/kk_BT) - 1)$. [e] The meta and ortho positions in the *p*-nitrobenzyloxy function.

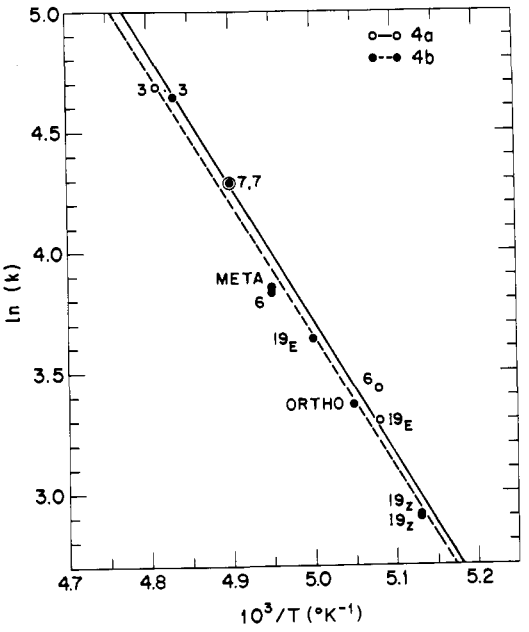


Figure 5. The plot of rate constants at coalescence (k_c) vs. the coalescence temperatures ($1/T$) for the various protons of (a) 4,4-dimethylvitamin D_3 (**4a**) (open circles) and (b) its *p*-nitrobenzoate ester (**4b**) (full circles).

of activation ($\Delta G_c^{\ddagger}$) at coalescence is obtained:

$$\Delta G_c^{\ddagger} = T_c(45.67 + 4.58 \log T_c/\Delta\nu) \quad (1)$$

where T_c is the coalescence temperature and $\Delta\nu$ is the chemical shift difference between the two sites at the slow exchange limit in hertz. Equation 1 will be valid provided that the population difference is not too large. It was shown by Kost and Raban[15] that when the chemical shift difference between the two signals is greater than ca. 5–10 Hz, the use of the above approximate relation for k_c is well justified.

We have also utilized the Arrhenius relation:[16]

$$\ln k = -\frac{E_a}{R}\left(\frac{1}{T}\right) + \ln A \quad (2)$$

where k is the rate constant; R is the gas constant in calories; E_a is the energy of activation; T is the temperature in K; and the usual intercept of ln A. The practice is to get a plot of ln k vs. $1/T$ for each proton at various temperatures, covering both the fast and slow exchange limits, but the most sensitive and thus the most accurate range of temperatures is the intermediate range just above and below the coalescence temperature. The plot is usually accomplished by computer-simulated spectra and visual comparison with the experimental spectra.[16] We have found that rather than obtaining a single plot for each hydrogen multiplet, we could obtain the same information by plotting the rate constants vs. the coalescence temperatures of all the hydrogen multiplets involved. This procedure is justified if only one process is involved, such as in this case the chair–chair interconversion. Under such a condition the individual proton plots must coincide (Figure 5). Since the coalescence temperatures for all multiplets cover the intermediate range of exchange (e.g., k_c = 120–18 s^{-1}), the sensitivity of this plot is comparable with that of the classical plot obtainable with the aid of computer-simulated spectra. The disadvantage of employing this procedure arises from the accuracy in determining the position of the coalescence temperatures. However, the error in determining the coalescence temperatures is not greater than ±2 K, which is, in fact, comparable with the error involved in measuring the true temperature of the sample using a chemical thermometer (CH_3OH). In order to reduce the errors involved in calculating the rate constants, we have used a simple computer Lorenzian curve fitting for those cases where the multiplets at the slow exchange limit have not been fully separated.

Table IV. Thermodynamic Data and Parameters for 1α-Hydroxy-4,4-dimethylvitamin D_3 (**5**) and 1α-Hydroxy-4,4-dimethylepivitamin D_3 (**6**)[a]

compd	signal obsd for protons at	T_c, ±2 K	k_c at T_c, s^{-1}	$\Delta G_c^{\ddagger}$ at T_c, kcal/mol	av $\Delta G_c^{\ddagger}$
5	C_1	226	127	10.94	10.95 ± 0.2
	C_2	220	60	10.96	
6	C_3	246	160	11.84	12.00 ± 0.2
	$C_{19\,(Z)}$	237	31	12.16	

[a] See footnotes to Table III.

In Table III we have collected all activation and thermodynamic parameters as evaluated by both methods.

Since the $\Delta G_c^{\ddagger}$ values obtained at T_c for the different protons in **4a** and **4b** are nonuniformly spread, and the $\Delta S_c^{\ddagger}$ values are small and within the experimental error, we may conclude that the $\Delta G_c^{\ddagger}$ values are temperature independent. This conclusion is supported by the excellent agreement between the

Table V. ^{13}C Chemical Shift Data for Vitamin D_3 (**1a**), 4,4-Dimethylvitamin D_3 (**18**), 4,4-Dimethylcholesterol (**19**), and 4,4-Dimethylepicholesterol (**19**)[a]

Carbon	1a	4a ($\Delta\delta$)[c]	18 ($\Delta\delta$)[c]	19
1	31.95	30.35[b] (−0.25)	36.75 (−0.35)	31.90
2	35.25	32.00[b] (−2.60)	27.50 (−3.65)	25.25
3	69.25	77.05 (3.55)	77.60 (2.0)	76.60
4	46.00	43.15 (−1.05)	41.65 (−0.25)	40.65
5	135.15[d]	144.75[b,e] (−0.9)	149.95 (−0.85)	147.40
6	122.50	118.75[b,e] (−0.3)	120.25 (0.5)	121.90
7	117.65	118.25[b] (−0.1)	32.65	32.60
8	142.30	142.10 (0.55)	30.95	30.20
9	29.05	29.10	51.05 (−0.15)	50.90
10	145.20[d]	144.50[e] (−)[f]	36.80 (−0.05)	36.50
11	22.30	22.35	20.65	20.50
12	40.60	40.55	39.85	39.80
13	45.90	45.85	42.30	42.15
14	56.40	56.45	57.35	57.30
15	23.60	23.50	24.25	24.20
16	27.65	27.60	28.30	28.30
17	56.75	56.60	56.20	56.10
18	12.05	12.00	11.90	11.90
19	112.40	113.60 (0.75)	21.35 (0.0)	21.85
20	36.15	36.00	35.85	35.80
4α		25.40[b] (−0.1)	27.25 (−0.05)	21.15
4β		22.10[b] (0.6)	23.65 (1.35)	28.05

[a] Shifts are given relative to Me_4Si and are accurate to within ±0.05 ppm. The side chain resonances not listed in the table are C_{21}, 18.85; C_{22}, 36.20; C_{23}, 23.90; C_{24}, 39.55; C_{25}, 28.05; C_{26}, 22.60; C_{27}, 22.85. [b] These signals were selectively broadened at −30 °C. [c] Values in parentheses are $\Delta\delta = \delta$ (ester) − δ (OH); any $\Delta\delta < 0.1$ ppm was neglected for all carbons other than ring A carbon atoms. [d] Original assignments have been reversed (ref 17). [e] Assignment down any column may be reversed. [f] This signal was not observed.

$\Delta G_c^{\ddagger}$ values (Table III) obtained with the aid of the approximate method (eq 1) and that obtained from the Arrhenius plot (eq 2).

It must be noted that unless $\Delta G°$ is zero, the $\Delta G_c^{\ddagger}$ values obtained by eq 1 represent the energy barrier for the less stable conformation of the two. Thus, if the equilibrium population ratio is near unity, which is practically true for **4a** and **4b,** and $\Delta G_c^{\ddagger}$ is a temperature-independent parameter, one would expect to obtain the same $\Delta G_c^{\ddagger}$ value using both methods.

The agreement in $\Delta G_c^{\ddagger}$, as obtained by both methods (Table III), justifies the use of the approximate method for this family of compounds.

b. ^1H NMR of 1α-Hydroxy-4,4-dimethylvitamin D_3 (5) and Its Epimer 6. In these two compounds, we have observed the slow exchange limit for only two hydrogen multiplets: H at C_1 and C_3 in **5,** H at C_3 and H_2 at C_{19} in **6**. Owing to the low solubility of these compounds we could not, however, obtain sufficiently low temperatures to observe the slow exchange limit for the other hydrogen multiplets. As a result, the thermodynamic data were obtained by the use of the approximate relation (eq 1) alone.

The chemical shift data at room and low temperatures are collected in Tables I and II, respectively, and the calculated activation parameters are shown in Table IV.

c. ^{13}C NMR of 4,4-Dimethylvitamin D_3 (4a), 4,4-Dimethylcholesterol (18), and Its C_3 Epimer 19. The ^{13}C chemical shift data and the assignment for 4,4-dimethylvitamin D_3 (**4a**), the *p*-nitrobenzoate ester **4b,** and the two model compounds 4,4′-dimethylcholesterol (**18**) and its C_3 epimer **19,** as well as the revised assignment for vitamin D_3 (**1a**),[17] are listed in Table V.

Most of these assignments were done using single frequency off-resonance decoupling (SFORD), partially relaxed spectra (PRS), and comparison with analyzed spectra of compounds having related structures. The distinction between the signals due to the two methyl groups at C_4 in dimethylcholesterol (**18**) was based on the comparison with the ^{13}C spectrum of its nitrobenzoate ester. The signal which shifted on esterification was assigned to the α-methyl at C_4.[18] In the dimethylepicholesterol **19,** the methyl signal at C_4 with the shorter T_1 value (in the partially relaxed spectrum) was assigned to the α-methyl by virtue of its gauche interaction with OH at C_3.[19]

The above analysis for **4a** did not allow unequivocal distinction between the pairs of signals due to C_5–C_{10}, C_6–C_7, and C_1–C_2. The assignment of these signals was based on their different temperature-dependent behavior, since at the slow exchange limit they are expected to have a different chemical shift difference ($\Delta\nu$), and thus will have a distinctly different coalescence temperature. On lowering the temperature one of the signals in each pair broadened more than the other, and the broadened signals were assigned to C_5, C_6, and C_1. These assignments are based on the larger shift effect experienced by these carbon atoms in their respective conformers.[20] Thus, C_5 will experience a larger chemical shift effect ($\Delta\nu$) than C_{10} owing to the OH at C_3, and C_6 a larger effect than C_7 owing to the methyls at C_4. In the case of C_1 and C_2 one of the signals (at 30.35 ppm) was broadened at a slightly higher temperature (−25 vs. −30 °C), which may be due to the γ interaction of the former with the OH at C_3.

We have recorded the low-temperature ^{13}C spectra of **4a** and have observed at −90 °C separate C_3 signals for each of the two conformers, the chemical shift difference between them being 1.0 ± 0.2 ppm.

Discussion

The slight differences in magnitude for the $\Delta G_c^{\ddagger}$ values obtained at different coalescence temperatures, and their random distribution (as calculated from the modified Eyring's relation, eq 1), as well as the comparatively small $\Delta S^{\ddagger}$ values (obtained from Arrhenius plot, eq 2), show that in the ring inversion the free energy of activation, $\Delta G_c^{\ddagger}$, is temperature independent.

The similar population ratio of the OH-equatorial to the OH-axial conformers in both 4,4′-dimethylvitamin D_3 (**4a**) and its ester **4b** indicates similar ground-state free-energy differences ($\Delta G°$) between the two conformers in both compounds. The same relationship between the respective conformers of the free alcohols and their *p*-nitrobenzoate esters

Table VI. The Dimethyl Substituent Chemical Shift Effects for Vitamin D_3 (**1a**) Cholesterol and Epicholesterol[a]

$\Delta\delta$ carbon	δ (**4a**) − δ (**1a**)	δ (**18**) − δ (cholesterol)	δ (**19**) − δ (epicholesterol)
1	−1.6	−0.6	−1.4
2	−3.25	−4.2	−3.7
3	7.8	5.85	9.45
4	−2.8	−0.7	0.7
5	9.6	9.1	8.8
6	−3.4	−1.4	−2.15
7	0.6	0.7	0.55
8	−0.2	−1.0	−1.75
9		0.8	0.45
10	−0.7	0.25	−0.9
11		−0.5	−0.35
12			
13			−0.25
14		0.5	0.45
19	1.2	1.95	3.15

[a] The resonances due to carbons 15–18 and 20–27 were not affected upon the dimethyl substitution.

was previously observed in vitamin D_3 (**1a**) and some of its analogues.[5] In addition, the data in Table III indicate that the free energies of activation for **4a** and **4b** are the same.

Thus, the contribution of the ester group to the nonbonded interactions both in ground and excited states is very small indeed.

Our failure to observe any spectral changes for vitamin D_3 (**1a**), even below −100 °C, indicates that the energy barrier for this compound is likely to be lower than that of 4,4-dimethylvitamin D_3 (**4a**) (<8.5 kcal/mol).[21] It is very unlikely that this failure to observe the slow exchange limit for **1a** arises from the intrinsic small chemical shift differences ($\Delta\nu$) for respective protons in the two conformers, since the magnetic environment of most ring A protons is expected to be almost the same in both **1a** and **4a**. The comparatively low energy barrier ($\Delta G^{\ddagger}$) for ring A inversion in **1a** is not surprising as its ring A sp^2 carbon atoms are supposed to lower this barrier considerably with respect to a normal cyclohexane ring.[17] Although the $\Delta G^{\ddagger}$ for the parent 1,2-dimethylenecyclohexane was not yet established, the $\Delta G^{\ddagger}$ values for methylenecyclohexane and its 2,2-dimethyl derivatives are lower than those for the respective saturated cyclohexane derivatives (ca. 8.5 vs. 10.0 kcal/mol).[22]

The observed increase in the energy barrier, when the two methyl groups are introduced into ring A of vitamin D_3 (**1a**), is probably due to an unfavorable interaction between these methyls and the H and C_6. The existence of such interaction in the ground state of **4a** is evident from the large mutual γ-deshielding effect on C_4 and C_6 upon the dimethyl substitution at C_4.[23,24] Thus, the ^{13}C chemical shift differences for C_4 and C_6 between **4a** and **1a** are −2.8 and −3.4 ppm, respectively. The shift differences between the analogous C atoms in 4,4-dimethylcholesterol (**18**) and cholesterol are −0.7 and −1.4 ppm, and those between 4,4-dimethylepicholesterol (**19**) and epicholesterol are 0.7 and −2.15 ppm (Table VI). This interaction between the methyls at C_4 and H at C_6 produces a strain on the C_5–C_6 double bond, as evident from the UV spectrum of **18** and **19** which show both a blue shift of 5 nm and a considerable decrease in the ϵ value compared with vitamin D_3 (**1a**).

Other interactions between these methyl groups and the ring A substituents are not expected to have a substantial effect on the energy profile of the ring A inversion. It has been shown by Bernard et al.[22] that the introduction of methyl groups, in vicinal position to an exo methylene group, does not substantially change the barrier to inversion in methylenecyclohexane.

The comparison of the ^{13}C NMR spectra of **4a** with those of **18** and its epimer **19** on one hand, and vitamin D_3 (**1a**) on the other hand, suggests that the rest of ring A interactions in **4a** are about the same as in the parent molecule **1a** (Table V). Thus the dimethyl substitution chemical shift (SCS) at C_3 in **4a** was midway between the corresponding dimethyl SCS in **18** and **19** (Table VI).

In the ^1H NMR spectrum of **4a** there are indications that the dihedral angles in ring A protons are the same for both **4a** and **1a**. The vicinal spin–spin coupling constants of the proton at C_3 in **4a**, at room temperature, are similar to those of **1a** ($^3J_{trans}$ = 7.3, $^3J_{cis}$ = 3.7 Hz vs. $^3J_{trans}$ = 7.6, $^3J_{cis}$ = 3.8 Hz, respectively). The trans coupling constants may be regarded as representing a weighted average value of the two limiting trans coupling constants $^3J_{ax:ax}$ = 11.1 and $^3J_{eq:eq}$ = 2.7 Hz, derived from cyclohexanol.[25] Similarly, the cis coupling constants of **4a** and **1a** may be regarded as the average values of $^3J_{eq:ax}$ and $^3J_{ax:eq}$. We have, in addition, obtained from the slow exchange limit spectra of **4a** the true trans coupling constants for the OH-equatorial conformer ($^3J_{ax:ax}$ = 11.4 and $^3J_{eq:eq}$ = 4.3 ± 0.4 Hz).[26] This result indicates that ring A in 4,4-dimethylvitamin D_3 (**4a**), and thus also in vitamin **1a** and its analogues, exists in two genuine chair conformations as in the case of cyclohexanol.[30]

Comparison of the activation parameters of **4a** with those of its two hydroxylated analogues **5** and **6** shows that the introduction of OH group at C_1 in a 1,3-trans relation increases the barrier by ca. 1 kcal/mol, while the introduction of OH group in a 1,3-cis relation by 2 kcal/mol. The first increase may be ascribed to the substituent effect of the additional OH group, while the second increase arises from the energy necessary to break the 1,3-hydrogen bonding during the chair inversion.[6]

It is to be noted that similar population ratios of the two conformers (C_3-OH eq vs. C_3-OH ax) were observed for both vitamin D_3 (**1a**) and its dimethyl analogue **4a** (57:43 and 55:45), as well as those of the respective cis 1,3-diols **2b** and **6** (20:80 and 21:79). However, the population ratios of the trans 1,3-diols **1b** and **5** differ slightly, favoring in the former the C_3-OH axial conformer, and in the latter, the C_3-OH equatorial conformer (44:64 and 67:37, respectively).

Addition of another sp^2 ring carbon atom lowers the barrier to the ring inversion of the dimethylvitamin D_3 derivatives, as evident by the lack of temperature dependence of the ^1H NMR spectra of **7** and **8**, the two keto analogues of vitamin D_3. These two ketones (as their 4,4-dimethylhydroxyl analogues **4a, 5,** and **6**) exist in solution in the chair conformation as may be recognized from their UV spectra which show enhanced n–π^* transitions and new π–π^* transitions, due to coupling between the nonconjugated carbonyl and double bond chromophores.[27]

Recently, we have proposed a method for the conformational analysis of vitamin D_3 and its analogues in the fast exchange limit region using ^{13}C NMR spectroscopy.[5] The method is based on the assumption that the C_3 chemical shift difference, Δ, between the two ring A chair conformations is equal to the C_3 chemical shift difference observed between cholesterol and epicholesterol (Δ = 4.62 ppm). This assumption is verified in the case of 4,4-dimethylvitamin D_3 (**4a**).

The C_3 chemical shift difference between the respective conformers of **4a** at the slow exchange limit, Δ = 1.0 ppm, was found to be identical with that between the C_3 chemical shifts of 4,4-dimethylcholesterol (**18**) and its C_3 epimer **19**. The smaller Δ value for the dimethyl derivatives, as compared to the nonmethylated compounds (1.0 vs. 4.62 ppm), reflects the dimethyl substituent chemical shift (SCS) effect on C_3.

The derivation of Δ values, from model systems, for conformational analysis under fast equilibrium conditions (e.g., **18** and **19** for **4a**) is justified because the origin of the Δ value

arises from neighboring atoms in the immediate vicinity of the carbon atom under consideration and can be related to the slightly different steric interactions imposed on the C atom in the two conformations.[20] It seems that a reliable estimate of the Δ value can be obtained if the carbon atom under consideration has the same relations to its neighboring atoms as far as two bonds away (γ effect), in both the compound under investigation and the model compounds, as evident from the dimethyl SCS values for **4a** vs. **18** and **19** (Table VI).[28] The only carbon atom conforming with these relations in **4a, 18,** and **19** is C_3. Since the population ratio of the two ring A conformers in **4a** is ca. 1:1, the magnitude of the SCS effect on C_3 in this compound is, as expected, midway between that on the respective carbon atoms in **18** and **19**.

Experimental Section

^{1}H NMR spectra were recorded on a Bruker WH-270 spectrometer. Flip angles of ca. 90° were employed with 16K transform which gave ca. 0.1 Hz per data point for a 1900-Hz sweep width. Peak positions were determined by a software control and are considered to be accurate to within 6×10^{-4} ppm. The samples studied were dissolved in CCl_3F (5% w/w) containing CD_2Cl_2 to provide the lock. Me_4Si was used as an internal reference. Temperatures were monitored by means of a Bruker temperature control unit, Model B-ST 100/700, and determined from the peaks separation of a calibrated methanol sample, and are considered accurate to ±2 °C. The ^{13}C NMR spectra were recorded on a Bruker WH-90 spectrometer, operating at 22.63 MHz. Samples were dissolved in $CDCl_3$ (ca. 0.05–0.2 M) containing some Me_4Si as an internal reference. Flip angles of ca. 60–70° were employed with 8K transform which gave ca. 0.5 Hz per data point for a 4000-Hz sweep width. Peak positions were determined by a software control and are considered accurate to within 0.05 ppm.

The low-temperature ^{13}C spectra of 4,4-dimethylvitamin D_3 (**4a**) were recorded on a Bruker WH-270 spectrometer operating at 67.89 MHz. Flip angles of ca. 45° were employed with 16K transform which gave ca. 1.5 Hz per data point for a 12 000-Hz sweep width. A solution of ca. 0.05 M **4a** in a 4:1 mixture of CCl_3F/CD_2Cl_2 containing Me_4Si as an internal reference was placed in a 10-mm NMR tube. Temperatures were monitored by means of a Bruker temperature control unit, Model B-ST 100/700, and determined from the peak separation of a calibrated MeI/Me_4Si (1:3) sample.[29]

The ultraviolet spectra were taken on a Cary 118 spectrophotometer. Mass spectra were recorded on a Varian MAT 731 high-resolution mass spectrometer.

4,4-Dimethylprevitamin D_3 (10). A solution of 500 mg of 4,4-dimethylcholesta-5,7-dien-3-ol (**9**)[9b] in 500 mL of ether was irradiated for 30 min with 300-nm light under nitrogen at 0 °C, the solvent was evaporated to dryness, and the residue was chromatographed on a silica gel dry column using hexane–ether (7:3) as an eluent resulting in 160 mg of previtamin **10**: λ_{max} (ether) 257 mm (ε 8000); on addition of I_2, λ_{max} 256 nm (ε 12 300); δ (CCl_4) 0.72 (s, 3, CH_3 + C_{13}), 1.67 (s, 3, CH_3 at C_{10}) 3.85 (quintet, J = 9.4 Hz, H-C_3), 6.13 (m, 3, H-C_6, -C_7, and -C_9).

Anal. ($C_{29}H_{48}O$). Found: *m/e* 412.3608.

4,4-Dimethylvitamin D_3 (4a). A solution of 100 mg of previtamin **10** in 50 mL of isooctane was heated for 4 h at 80 °C. The solvent was then evaporated and the residue was chromatographed on a silica gel column using 7:3 hexane–ether as an eluent to give 80 mg of **4a**: λ_{max} 261 nm (ε 12 000); NMR, Table I.

Anal. ($C_{29}H_{48}O$). Found: *m/e* 412.3602.

4,4-Dimethylvitamin D_3 *p*-Nitrobenzoate (4b). A solution of 50 mg of **4a** in 3 mL of pyridine was treated with 50 mg of *p*-nitrobenzoyl chloride and stirred overnight at room temperature. Pyridine was evaporated to dryness and the residue was chromatographed on a silica gel column. Elution with hexane–ether (8:2) gave 45 mg of the *p*-nitrobenzoate, **4b**: mp 167–169 °C; λ_{max} (ether) 256 nm (ε 22 000); NMR, Table I.

4,4-Dimethyl Ketone 7. A solution of 100 mg of **4a** in 3 mL of dimethyl sulfoxide was treated consecutively with 50 mg of diethylcarbodiimide, 1 mL of pyridine, and 0.5 mL of trifluoroacetic acid and then stirred for 8 h at room temperature. The material was eluted with ether and chromatographed on a dry silica gel column. Elution with hexane–ether (8:2) gave 80 mg of the ketone **7**: λ_{max}, see text; δ (CCl_4) 0.55 (s, 3, CH_3-C_{13}), 4.93, 5.21 (m, 2, H-C_{19}), and 5.95, 6.25 (AB_q, J = 11 Hz, 2, H-C_6 and H-C_7).

Anal. ($C_{29}H_{46}O$). Found: *m/e* 410.3448.

4,4-Dimethylcholesta-1,5,7-trien-3-one (13). A solution of 1 g of cholesta-1,5,7-trien-3-one (**12**) in 50 mL of dry benzene was added to a solution of 2 g of sodium methoxide in 100 mL of dimethyl sulfoxide at room temperature, under nitrogen. The solution was stirred for 10 min, then 10 g of methyl iodide was added and the stirring was continued for an additional 1 h. The product was extracted with 500 mL of ether. The ether extract was washed with brine, dried, and evaporated to dryness to give 0.7 g of **13**: mp 64–65 °C (methanol); $(\alpha)_D$ −46° (dioxane); λ_{max} (ether) 275 nm (ε 6000); δ ($CDCl_3$) 0.75 (s, 3, CH_3-C_{13}), 5.8 and 5.7 (AB_q, J = 5.5 Hz, H-C_6 and H-C_7).

1α,2α-Epoxy-4,4-dimethylcholesta-5,7-dien-3-one (14). A solution of 5% potassium hydroxide in 10 mL of methanol and 5 mL of 50% hydrogen peroxide were added to a solution of 1 g of 4,4-dimethylcholesta-1,5,7-trien-3-one (**13**). The reaction mixture was stirred overnight at room temperature under nitrogen and the product was filtered off, washed with water, and dried under vacuum. Crystallization from ether–hexane gave 0.8 g of **14**: mp 155–156 °C (hexane); $(\alpha)_D$ +24° (dioxane); λ_{max} (ether), 293 s, 282, 272, 261 s nm (ε 5400, 9300, 9300, 7000); δ (CCl_4) 0.55 (s, CH_3-C_{13}), 3.4 and 3.5 (AB_q, J = 2 Hz, 2, H-C_1 and H-C_2), 5.4 and 5.8 (AB_q, J = 6 Hz, H-C_6 and H-C_7).

4,4-Dimethylcholesta-5,7-diene-1α,3β-diol (15a) and 4,4-Dimethylcholesta-5,7-diene-1α,3α-diol (16a). A solution of 500 mg of 1α,2α-epoxy-4,4-dimethylcholesta-5,7-dien-3-one (**14**) in 50 mL of ether was added dropwise to a stirred mixture of 0.5 g of lithium aluminum hydride in 20 mL of dry ether. The reaction mixture was refluxed for 3 h and then was treated dropwise with a saturated solution of sodium sulfate. Solid sodium sulfate was added and then the mixture was filtered. The crude product isolated from ether was separated into two fractions by thin layer chromatography, on plates coated with silica gel developed with ether. One fraction gave 80 mg of 4,4-dimethylcholesta-5,7-diene-1α,3β-diol (**15a**): mp 159–160 °C (acetone); $(\alpha)_D$ −111° (dioxane, *c* 0.7); λ_{max} (ether) 293 s, 282, 272, 261 s nm (ε 7000, 12 000, 12 000, 8700); δ (CCl_4) 0.5 (s, 3, CH_3-C_{13}), 3.75 (m, 2, H-C_2), and 5.35, 5.90 (AB_q, J = 5.5 Hz, H-C_6 and H-C_7).

The other fraction contained 320 mg of 4,4-dimethylcholesta-5,7-diene-1α,3α-diol (**16a**): mp 180–182 °C (hexane); $(\alpha)_D$ −105° (dioxane); λ_{max} (ether) 292 s, 282, 272, 261 s nm (ε 6000, 11 000, 11 000, 8200); δ (CCl_4) 0.52 (s, 3, CH_3-C_{13}), 3.55 (m, 2, H-C_1 and H-C_3), 5.78, 5.83 (AB_q, J = 10 Hz, H-C_6 and H-C_7).

4,4-Dimethylcholesta-5,7-diene-1α,3β-diol 3-Acetate (15b). A solution of 100 mg of **15a** was dissolved in 1 mL of pyridine and treated with 0.5 M acetic anhydride. Isolation from ether and purification on a thin layer plate coated with silica gel developed with ether hexane (1:1) yielded 75 mg of **15b**: mp 182–183 °C (acetone); λ_{max} (ether) 293 s, 282, 272, and 261 s nm (ε 4600, 9300, 9300, 7000); δ (CCl_4) 0.52 (s, 3, CH_3-C_{13}), 2.0 (s, 3, H-OAc), 3.7 (m, 1, H-C_1), 5.04 (t, J = 8 Hz, 1, H-C_3), 5.80, 8.30 (AB_q, J = 11 Hz, H-C_6 and H-C_7).

4,4-Dimethylcholesta-5,7-diene-1α,3α-diol 3-Acetate (16b). A solution of 75 mg of **16a** in 10 mL of methylene chloride was treated with 220 mg of dimethylaminopyridine and 0.3 mL of acetic anhydride overnight at room temperature. The product was isolated from ether and purified on a preparative thin layer silica gel plate developed with hexane–ether (1:1) to give 54 mg of **16b**: mp 136–138 °C (hexane); λ_{max} (ether) 293 s, 282, 272, and 261 s nm (ε 5000, 9000, 9000, 7000); δ (CCl_4) 0.53 (s, 3, CH_3-C_{13}), 3.50 (m, 1, H-C_1), 4.9 (m, 1, H-C_3), 5.78 and 5.84 (AB_q, J = 10 Hz, H-C_6 and H-C_7).

1α-Hydroxy-4,4-dimethylcholesta-5,7-dien-3-one (17). A solution of 100 mg of **16a** in 1 mL of dimethyl sulfoxide and 0.5 mL of dry benzene was treated with 0.05 mL of pyridine, 0.05 mL of trifluoroacetic acid, and 1 mL of diethylcarbodiimide. After 2 h the product was isolated from ether and chromatographed on thin layer plates coated with silica gel, with ether–hexane (1:1) to give 50 mg of **17**: mp 185–186 °C (hexane); $(\alpha)_D$ −66° (dioxane, *c* 1); λ_{max} (ether) 293 s, 282, 272, 261 s nm (ε 5500, 9600, 9600, 7400); δ (CCl_4) 0.55 (s, 3, CH_3-C_{13}), 3.90 (m, 1, H-C_1), 5.42 (m, 1, H-C_7), 5.74 (d, J = 5.5 Hz, H-C_6).

Reduction of 1α-Hydroxy-4,4-dimethylcholesta-5,7-dien-3-one (17). A solution of 100 mg of **17** in ether was added to a mixture of 50 mg of lithium aluminum hydride in ether and stirred for 30 min at room temperature; the material was isolated from ether and chromatographed on silica gel coated thin layer plates with ether to give 30 mg

of **15a** and 30 mg of **16a**, identical with the samples described above.

1α,2α-Epoxy-4,4-dimethylcholesta-5,7-dien-3α-ol and Its Reduction to 16a. A solution of 200 mg of **14** in 15 mL of dry ether was added dropwise to a slurry of lithium aluminum hydride in 10 mL of ether at 0 °C. After the solution was stirred for 30 min at this temperature, a saturated sodium sulfate solution was added and the isolated material was separated on silica gel plates to give 75 mg of the title compound: mp 168–169 °C; λ_{max} (ether) 293 s, 282, 272, and 261 s (ε 6000, 11 000, 11 000, 8000); δ (CCl_4) 0.55 (s, 3, CH_3-C_{13}), 0.77 (s, 3, CH_3-C_{19}), 2.98 (s, 1, H-C_1), 3.40, 3.48 (AB_q, J = 2 Hz, H-C_1 and H-C_2), 5.41 (m, 1, H-C_7), 5.62 (d, J = 5.2 Hz, H-C_6).

The solution of 75 mg of epoxy alcohol in 10 mL of ether was added to a slurry of lithium aluminum hydride in 5 mL of ether. The product was isolated as above to give 55 mg of 4,4-dimethylcholesta-5,7-diene-1α,3α-diol (**16a**), mp 180–181 °C, identical with the material obtained above.

4,4-Dimethyl-1α-hydroxyvitamin D_3 (5). A solution of 100 mg of **15a** in 300 mL of ether was irradiated for 30 min with 300-nm light which was filtered with 0.4% sodium nitrate solution, at 0 °C and under nitrogen atmosphere. The solvent was then evaporated under vacuum at 0 °C, the residue dissolved in 50 mL of isooctane, and the solution heated at 80 °C for 4 h under nitrogen atmosphere. The solvent was evaporated and the residue was chromatographed on a silica gel column. Elution with ether gave 30 mg of **5**: mp 149–150 °C (acetone); $(\alpha)_D$ +8° (ether, c 0.5); λ_{max} (ether) 257 nm (ε 11 500); NMR, see Table II.

Anal. ($C_{29}H_{48}O_2$). Found: *m/e* 428.3605.

4,4-Dimethyl-1α-hydroxy-3-epivitamin D_3 (6). 16a (100 mg) was irradiated as described above to give 30 mg of **6**: mp 116–118 °C (acetone); $(\alpha)_D$ +19° (ether); λ_{max} (ether) 257 nm (ε 11 300); NMR, see Table II.

Anal. ($C_{29}H_{48}O_2$). Found: *m/e* 428.3601.

3-Keto Analogue of 4,4-Dimethyl-1α-hydroxyvitamin D_3 (8). The ketol **17** (50 mg) was irradiated as described above to give 10 mg of the ketone **8**: λ_{max} (ether) 320 s, 310 s, 290 s, 284, and 253 nm (ε 1200, 2700, 4000, 12 000, and 10 800); δ (CCl_4) 0.49 (s, 3, CH_3-C_{13}), 4.25 (d, d, J = 6.3 and 7.1 Hz, H-C_3) 4.96 and 5.45 (m, 2, H-C_{19}), 5.86, 6.28 (AB_q, J = 11.5 Hz, H-C_6 and H-C_7).

References and Notes

(1) G. M. Sanders, J. Pot, and E. Havinga, *Fortschr. Chem. Org. Naturst.*, **27**, 131 (1969); E. Havinga, *Experientia*, **29**, 1181 (1973).
(2) G. N. La Mar and D. L. Budd, *J. Am. Chem. Soc.*, **96**, 7317 (1974).
(3) R. M. Wing, W. H. Okamura, M. R. Pirio, S. M. Sine, and A. W. Norman, *Science*, **186**, 939 (1974); R. M. Wing, W. H. Okamura, A. Rego, M. R. Pirio, and A. W. Norman, *J. Am. Chem. Soc.*, **97**, 4980 (1975); W. H. Okamura, M. L. Hammond, A. Rego, A. W. Norman, and R. M. Wing, *J. Org. Chem.*, **42**, 2284 (1977).
(4) K. Rsukida, K. Akutsu, and K. Saiki, *J. Nutr. Sci. Vitaminol.*, **21**, 411 (1975).
(5) E. Berman, Z. Luz, Y. Mazur, and M. Sheves, *J. Org. Chem.*, **42**, 3325 (1977).
(6) M. Sheves, E. Berman, D. Freeman and Y. Mazur, *J. Chem. Soc., Chem. Commun.*, 643 (1975); W. H. Okamura and M. R. Pirio, *Tetrahedron Lett.*, 4317 (1975).
(7) M. Sheves, N. Friedman, and Y. Mazur, *J. Org. Chem.*, **42**, 3597 (1977).
(8) W. H. Okamura, A. W. Norman, and R. M. Wing, *Proc. Natl. Acad. Sci. U.S.A.*, **71**, 4194 (1974); W. H. Okamura, M. N. Mitra, D. A. Procsal, and A. W. Norman, *Biochem. Biophys. Res. Commun.*, **65**, 24 (1975).
(9) (a) Ohki, *Chem. Pharm. Bull.*, **8**, 46 (1960); (b) G. Cooley, B. Ellis, and V. Petrow, *J. Chem. Soc.*, 2998 (1955).
(10) W. H. Okamura, M. L. Hammond, H. J. C. Jacobs, and J.-V. Thuijl, *Tetrahedron Lett.*, 4807 (1976).
(11) K. E. Pfitzner and J. G. Moffat, *J. Am. Chem. Soc.*, **85**, 3027 (1963). Dicyclohexylcarbodiimide was replaced by diethylcarbodiimide (private communication by N. M. Weinshenker).
(12) After this work was finished, a publication by J. Brynjolffsen, J. M. Midgley, and W. B. Whalley appeared in *J. Chem. Soc., Perkin Trans. 1*, 812 (1975), dealing with the synthesis of the two diols **15a** and **18a** by a related route.
(13) E. Glotter, M. Weissenberg, and D. Lavie, *Tetrahedron*, **26**, 3857 (1970).
(14) Reduction of the epoxy ketone **14** with $LiAlH_4$ at 0 °C resulted in 1α,2α-epoxy-4,4-dimethylcholesta-5,7-dien-3α-ol which on treatment with the same reagent in boiling ether gave only the 1α,3α-diol **16a**.
(15) D. Kost, E. H. Carlson, and M. Raban, *Chem. Commun.*, 656 (1971).
(16) G. Binsch, *Top. Stereochem.*, **3**, 121 (1969).
(17) The assignment of the two signals to C_5 and C_{10} was reversed from our previous assignment (ref 5), on the basis of lanthanide-induced shifts experiment using Yb(fod)$_3$ and is in agreement with that in ref 4 and comparable with the assignment of the respective signals in **4a**.
(18) K. Tori, S. Seo, A. Shinaoka, and Y. Tomita, *Tetrahedron Lett.*, 4227 (1974); K. Tori, Y. Sakurawi, Y. Tomita, and H. Ishii, *ibid.*, 4163 (1976).
(19) E. Breitmaier, K. Spohn, and S. Berger, *Angew. Chem., Int. Ed. Engl.*, **14**, 144 (1975).
(20) J. B. Stothers, "^{13}C-NMR Spectroscopy", Academic Press, New York, N.Y., 1972, p 420.
(21) The upper limiting value of $\Delta G^{\ddagger}$ for vitamin D_3 (**1a**) is estimated from the data given for dimethylvitamin D_3 (**4a**) (where $\Delta\delta(C_3)$ = 1 ppm, $E_a \simeq$ 11.0 kcal/mol, $T_c \simeq$ 190 K) and the fact that at ca. 170 K no spectral changes are observed for the C_3 signal of **1a** ($\Delta\delta(C_3)$ = 4.62 ppm, see ref 5). The ratio between the C_3 chemical shift differences ($\Delta\delta$) for the two compounds, as a first approximation, equals the ratio of the Boltzmann factors of the rate constants.
(22) M. Bernard, L. Canuel, and M. St. Jacques, *J. Am. Chem. Soc.*, **96**, 2929 (1974), and references cited therein.
(23) S. H. Grover and J. B. Stothers, *Can. J. Chem.*, **53**, 589 (1975).
(24) G. E. Maciel, "Topics in ^{13}C-NMR Spectroscopy", Vol. 1, G. C. Levy, Ed., Wiley, New York, N.Y., 1974, p 64.
(25) F. A. L. Anet, *J. Am. Chem. Soc.*, **84**, 1053 (1962).
(26) It is possible to calculate the population ratio of **4a** at room temperature using the observed limiting coupling constants, as evaluated from its low-temperature spectrum. The relation used is $^3J_{obsd} = {}^3J_{a:a} \times P + {}^3J_{e:e} \times (1 - P)$, where P is the fractional population of the equatorial conformer.[2] The calculated population ratio using this method is 42:58 (eq-OH/ax-OH), which differs slightly from the result obtained using the limiting coupling constants derived from cyclohexanol[19] (55:45). The experimental error in the former case is ca. 17% while in the second case it is estimated to be 10%. This discrepancy between the two calculated population ratio points out the possible drawback in using model systems. Nevertheless, the deviations are small enough to exclude the possibility of puckered chair conformation.
(27) Half-chair or boat conformations of ring A in **7** or **8** are not expected to show such UV interactions. Cf. H. Labhart and G. Wagniere, *Helv. Chim. Acta*, **42**, 2219 (1959); M. Gorodetsky, A. Yogev, and Y. Mazur, *J. Am. Chem. Soc.*, **88**, 699 (1966).
(28) The smaller SCS values for C_6 and C_{19} in **4a**, as compared with **18** and **19**, are presumably due to the fact that the former are sp^2-hybridized carbon atoms. The greater deshielding effect of C_4 and C_6 in **4a** is due to an additional steric interaction between the methine proton at C_6 and the methyl groups at C_4. Smaller SCS deviations are observed for C_1 (ca. 0.6 ppm), C_2 (ca. 0.7 ppm), and C_5 (ca. 0.7 ppm) in **4a**.
(29) D. W. Vidrine and P. E. Peterson, *Anal. Chem.*, **48**, 1301 (1976).
(30) This is consistent with previous results of La Mar[2] and Okamura[3] from ^{1}H NMR conformational analysis at room temperature.