Tetrahedron Vol. 40, No. 7, pp. 1179 to 1182, 1984
Printed in Great Britain.

0040-4020/84 $3.00 + .00
Pergamon Press Ltd.

1α-HYDROXY PREVITAMIN D_3 AND ITS SELECTIVE FORMATION FROM 1-KETO PREVITAMIN D_3

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(*Received in UK* 15 *December* 1983)

Summary - An efficient method for the preparation of crystalline 1α-hydroxy previtamin D_3 is described. Also the stereoselective reduction of 1-keto previtamin 9 is discussed; it was observed that with aluminum hydride 7 is the most abundant product. This stands in contrast with previously reported $LiAlH_4$ and $NaBH_4$ mediated reductions.

The recognition[1] of the importance of 1α-hydroxylated vitamin D_3 metabolites and analogs for mediating intestinal calcium absorption and bone calcium mobilization has stimulated the study of various approaches to their synthesis[2]. However little attention so far has been devoted to practical syntheses of 1α-hydroxylated previtamin D_3 (7), the fundamentally important minor isomer in the natural equilibrium vitamin D ⇌ previtamin D (20 % previtamin at 36°C). Among the existing methods leading to 1α-hydroxy vitamin D_3, only the classical fotochemical-thermal isomerization of 1α-hydroxy provitamin D_3 provides a direct access to 1α-hydroxy previtamin D_3 (7)[3,4,5].

One important reason for having good access to 1α-hydroxy previtamin D_3 (7) is related to the observation of Mazur et al.[4] indicating that of the two isomers 6 and 7 the latter is the better substrate for allylic oxidation[4,6] to 1-keto previtamin D_3 (9). The reduction of 9 has been reported to yield predominantly (vide infra) 1β-hydroxy vitamin D_3 (after thermal isomerization), which is a serious drawback with regard to eventual preparation of radiolabelled 1α-hydroxy vitamin D_3.

In this context we wish to report on a modification of our previously described synthesis[7] of 6 (1 → 3 → 4 → 5) which leads conveniently to crystalline 1α-hydroxy previtamin D_3 (7) and on the stereoselective reduction of 9 to 7 (and hence to 6).

Of crucial importance in our approach is the possibility of adequate protection, as a Diels-Alder adduct, of the 6,8 diene in previtamin D_3 (1)(obtained either from vitamin D_3 by equilibration or from provitamin D_3 by low temperature irradiation). This allows selective carbonyl introduction at C-1 affording the triazoline adduct 4, followed by stereoselective reduction to 5. The subsequent one pot (15 N KOH-MeOH, reflux, 70 hr, Ar) generation of the vitamin triene system (cf. 6) almost necessarily has to involve four discrete transformations : (1) saponification to the cyclic hydrazine 10; (2) oxidation to the elusive unsaturated azo derivative[8]; (3) facile stereospecific cycloreversion to the original previtamin skeleton 7; (4) thermal isomerization to 6 under the conditions used. In view of this scheme, however, the direct obtention of 6 merely implies that either the oxidation step (2) occurs in spite of the "inert" atmosphere or that another pathway is in fact involved[8,9]. If the above cited sequence is operative it is obvious that when the reaction at higher temperature can be stopped at the stage of 10, subsequent low temperature oxidation would lead exclusively to 1α-hydroxy

previtamin D_3 (7). Reinvestigation of the reaction conditions carried out as a model study, on 3, has revealed that indeed traces of oxygen were present.

We have now found that when the hydrolysis is carried out under enforced oxygen-free conditions (vigorous reflux in carefully degassed 15 N KOH-MeOH, under fast Ar flow) the hydrazine 11 is indeed an intermediate which can be detected after rapid work-up. The highly air sensitive 11 was difficult to isolate; it was characterized as the dibenzoyl derivative 12

Consequently, treatment of 5 (as described for 3) followed by aerial oxidation of crude 10 (diethyl ether, air flow, 0°C) leads after recrystallization (diethyl ether) in 74 % to the desired 1α-hydroxy previtamin D_3 (7; m.p. 120-122°C).

Oxidation of 7 (commercial MnO_2 in diethyl ether, Argon, ultrasonic bath) gave (67 %) 1-keto previtamin 9. With 9 in hand we investigated the possibility for the stereoselective reduction to 7 using internal hydride delivery which proved successful in the transformation of 4 to 5. Treatment of 9 with aluminum hydride (prepared in situ) in THF at -60°C yielded a mixture of 7 and 8 (ratio 4:1; 91 % total yield) from which 7 was obtained in 72 % yield by preparative HPLC (Waters Model : 6000 A Solvent Delivery System HPLC; hexane/aceton; 9:1). This stands in contrast with previously described results which have shown the opposite stereoselectivity; a comparison is given in the table.

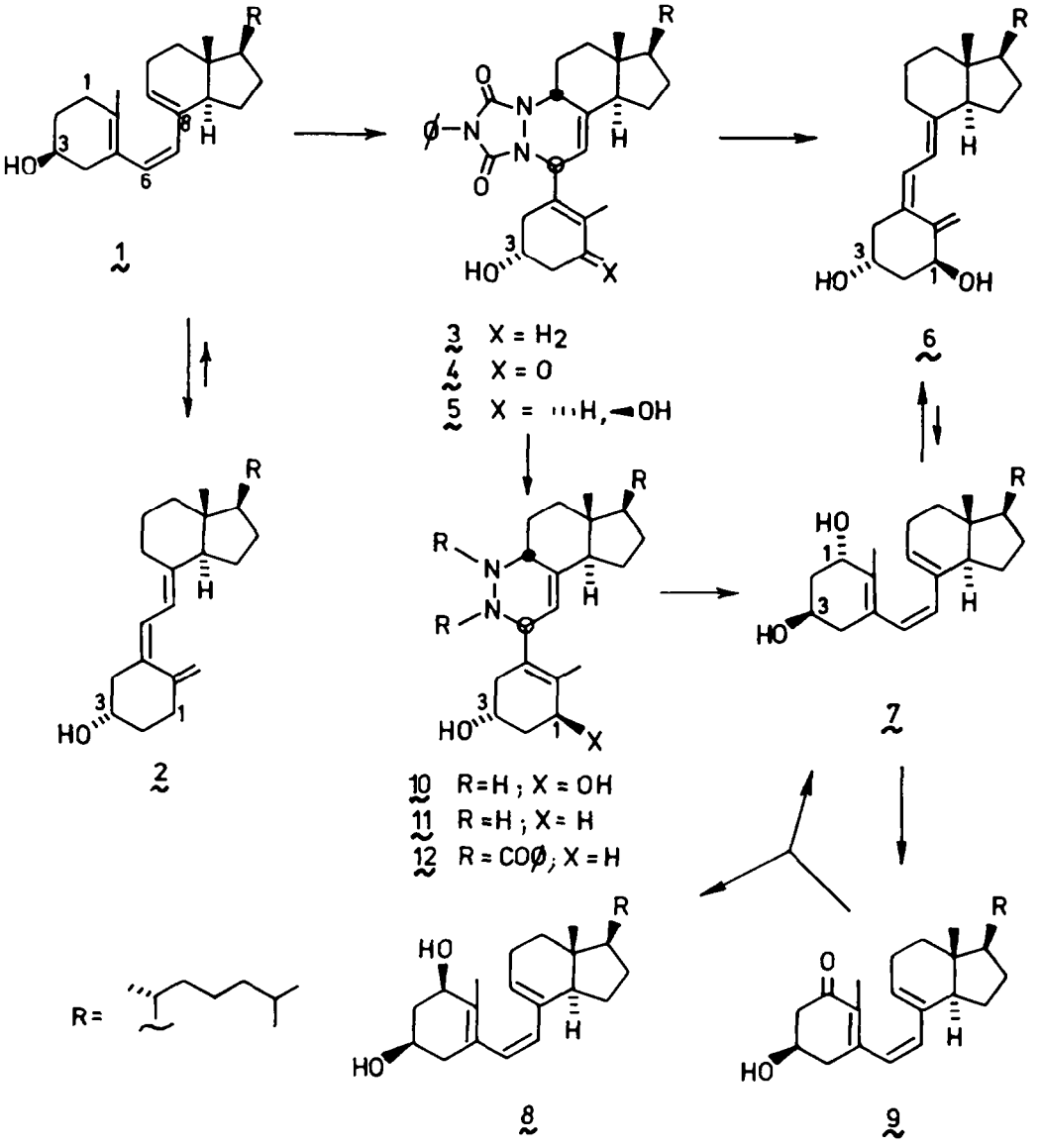

Table : Observed stereoselectivity upon reduction of 9

Reagent	7 (1α-OH)	:	8 (1β-OH)	Yield %	Ref.
$NaBH_4$, CH_3OH 0°C	0		1	70	4
$LiAlH_4$, ether, 0°C	1		2.8	60	4
$LiAlH_4$*, ether, -20°C	1		4	100	6
AlH_3, THF, -60°C	4		1	91	

* inverse addition

Thus, when aluminum hydride is the reducing agent, faster interaction between the 3-hydroxyl group and the aluminum atom assures internal hydride introduction at C-1.

Because of the obvious advantage of introducing the label in the last step in the synthesis of a radioactive compound our result offers the possibility for a suitable preparation of 1β-tritium labelled 1α-hydroxylated vitamin D_3.

EXPERIMENTAL SECTION

The m.ps. are uncorrected. IR spectra were recorded on a Beckman IR 4230 spectrophotometer, UV spectra on a Unicam SP 1750 Ultraviolet Spectrophotometer and mass spectra on a AEI MS-50 spectrometer. The ^1H NMR spectra were recorded at 360 MHz (WH-Brucker) in $CDCl_3$ unless otherwise stated with TMS as internal standard. Chemical shifts (δ) are expressed in ppm. Rf values are quoted for Merck silica gel 60 GF_{254} TLC plates of thickness 0.25 mm.

Reaction products were isolated by the addition of water and extraction with diethyl ether. The combined extracts were washed with brine and dried over $MgSO_4$. The solvent was removed from the filtered solution on a rotary evaporator. Column chromatographic separations were performed on silica gel, on a Prep LC/System 500 (Waters) apparatus or on a Model 6000 A Solvent Delivery System HPLC (Waters).

Formation of the dibenzamide 12

A soln of KOH in degassed H_2O (21 ml; 112 g in 84 mL H_2O) was added dropwise to a soln of 3 (500 mg; 0.89 mmol) in degassed CH_3OH (21 mL) at 110°C. Oxygen-free argon was passed through the soln at 110°C for 70 h. After cooling, the soln was extracted under argon with CH_2Cl_2 (5 x 50 mL). After work-up under argon and evaporation, the residue was dissolved in dry Et_2O (4 mL) and pyridine (0.07 mL). Benzoyl chloride (0.1 mL) was added dropwise to the soln at 0°C. After stirring for 2 h an ice-water mixture was added. Extraction with Et_2O, work-up and column chromatography (HPLC; hexane/acetone; 85:15) afforded 12 (202 mg; 36 %). Vitamin D_3 (2), previtamin D_3 (1) and their benzoates were also formed. Rf (benzene/acetone; 9:1) = 0.25.

12 : UV (CH_3OH) : λ_{max} = 213 nm. IR (KBr) : 3420, 3050, 2940, 2920, 2860, 1680, 1635, 1600, 1580, 1490, 1460, 1440, 1370, 1330, 1280, 910 cm^{-1}. ^1H NMR : 0.880 (d, J = 6.5 Hz, 3), 0.882 (d, J = 6.5 Hz, 3), 0.90 (s, 3), 0.94 (d, J = 5 Hz, 3), 1.26 (s, 3), 3.72 (m, 1), 4.75 (m, 1), 5.19 (, 1), 5.61 (m, 1), 7.17 (m, 2), 7.30-7.44 (m, 6), 7.64 (m, 2). MS : m/z 622 ($M^{+\cdot}$, 9), 394 (63), 106 (44), 105 (100), 77 (100), 43 (58).

1(s)-Hydroxy previtamin D_3 (7)

From 1-keto previtamin D_3 9 :

A soln of $AlCl_3$ (128 mg; 0.942 mmol) in dry THF (3.2 mL) was added to a suspension of $LiAlH_4$ (108 mg; 2.826 mmol) in dry THF (10 mL) at ambient temp. The suspension was placed in an ultrasonic bath for ten min.
A soln of AlH_3 (0.942 mmol; 3.3 mL of the prep. soln) was added dropwise to a soln of 1-keto previtamin D_3 (9) (75 mg; 0.1884 mmol) in dry THF (0.8 mL) at -70°C. After stirring for an additional 30 min at -60°C 5 % HCl soln (2 mL) and THF (2 mL) were added. Extraction with diethylether (3 x 30 mL), work-up and column chromatography (HPLC; hexane/acetone; 8:2) afforded 7 (55 mg) and 1(R)-hydroxy previtamin D_3 (8) (14 mg). The total yield was 91.5 % and the ratio 1(S)/1(R) ∿ 4/1.

7 : m.p. = 120-122°C (Et_2O); Rf (benzene/acetone; 7:3) = 0.29. UV (CH_3OH) : λ_{min} = 234 nm, λ_{max} = 260 nm. IR (KBr) : 3500, 3040, 3000, 2980, 2960, 1650-1600, 1470, 1390 cm^{-1}. ^1H NMR 0.70 (s, 3H), 0.866 (d, J = 6.8 Hz, 3), 0.871 (d, J = 6.8 Hz, 3), 0.94 (d, J = 6.4 Hz, 3), 1.77 (s, 3), 4.05 (m, FWHM = 24 Hz, 1), 4.20 (m, FWHM = 9 Hz, 1), 5.50 (m, 1), 5.79 (d, J = 12 Hz, 1), 5.91 (d, J = 12 Hz, 1).

8 : Rf (benzene/acetone; 7:3) = 0.42. UV (CH_3OH) λ_{min} = 228 nm, λ_{max} = 256 nm. ^1H NMR 0.71 (s, 3), 0.867 (d, J = 6.5 Hz, 3), 0.872 (d, J = 6.5 Hz, 3), 0.94 (d, J = 6.5 Hz, 3), 1.81 (s, 3), 4.02 (m, FWHM = 10 Hz, 1), 4.24 (m, FWHM = 11 Hz, 1), 5.55 (m, 1), 5.79 (d, J = 12 Hz, 1), 5.95 (d, J = 12 Hz, 1).

From adduct 5 :

A soln of KOH in degassed H_2O (30 mL; 112 g in 84 mL H_2O) was added to a soln of 5 (600 mg; 1.0435 mmol) in degassed CH_3OH (30 mL) at 110°C. Oxygen-free argon was passed through the soln at 110°C and the reaction was monitored by TLC. After completion CH_3OH was removed upon destillation under argon. Et_2O (30 mL) and CH_2Cl_2 (20 mL) were added at 0°C and the water layer was extracted with Et_2O-CH_2Cl_2 (3:2; 5 x 40 mL). The combined organic layers were washed with brine until neutral. At 0°C air was passed through the soln for 24 h. Work-up and column chromatography (HPLC; hexane/acetone; 8:2) afforded 7 (230 mg; 55 %).

Acknowledgments : The "Nationaal Fonds voor Wetenschappelijk Onderzoek" (NFWO) and the "Ministerie voor Wetenschapsbeleid" are gratefully acknowledged for financial support to the laboratory. L.J.V., Research Fellow of the NFWO, and P.J.D.C., Research Associate of the NFWO, thank the Fund for their positions.

References

1. H.F. DeLuca, H.K. Schnoes, Annu. Rev. Biochem., 45, 631-666 (1976); P.E. Georghiou, Chem. Soc. Rev., 6, 83 (1977).

2. B. Lythgoe, Chem. Soc. Rev., 9, 449 (1980). H. Jones, G.H. Rasmusson, Fortschr. Chem. Org. Naturst., 39, 121 (1980).
3. D.H.R. Barton, R.H. Hesse, M.M. Pechet, E. Rizzardo, (a) J. Am. Chem. Soc., 95, 2748 (1973); (b) J.C.S. Chem. Comm., 203 (1974).
4. M. Sheves, N. Friedman, Y. Mazur, J. Org. Chem., 42, 3597 (1977).
5. W.G. Dauben, R.B. Phillips and P. Jefferies, Proc. Nath. Acad. Sci. USA, 79, 5115 (1982).
6. H.E. Paaren, H.K. Schnoes, H.F. DeLuca, J.C.S. Chem. Comm., 890 (1977).
7. L.J. Vanmaele, P.J. De Clercq, M.E. Vandewalle, Tetrahedron Lett., 23, 995 (1982).
8. This step is assumed in view of the much higher reactivity for cycloreversion of unsaturated azo compounds compared to the corresponding unsaturated hydrazo compounds; see : B.T. Gillis and P.E. Beck, J. Org. Chem., 28, 3177 (1963); J.A. Berson, S.S. Olin, E.W. Petrillo, Jr., P. Bickart, Tetrahedron, 30, 1639 (1974) and references cited therein.
9. W. Reischl, E. Zbiral, Liebigs Ann. Chem., 745 (1978); D.J. Aberhart, A. Chi-Tung Hsu, J. Org. Chem., 41, 2098 (1976).