Bioorganic & Medicinal Chemistry Letters, Vol.3, No.9, pp. 1821-1824, 1993
Printed in Great Britain

0960-894X/93 $6.00 + .00
Pergamon Press Ltd

TOTAL SYNTHESIS OF 1α,25,28-TRIHYDROXYERGOCALCIFEROL

Andrew D. Batcho, John F. Sereno, Bernard M. Hennessy, Enrico G. Baggiolini,[1] and Milan R. Uskoković
Roche Research Center, Hoffmann-La Roche Inc., Nutley, New Jersey 07110

Ronald L. Horst
U.S. Department of Agriculture, National Animal Disease Center, Ames, Iowa 50010

(*Received* 5 *April* 1993)

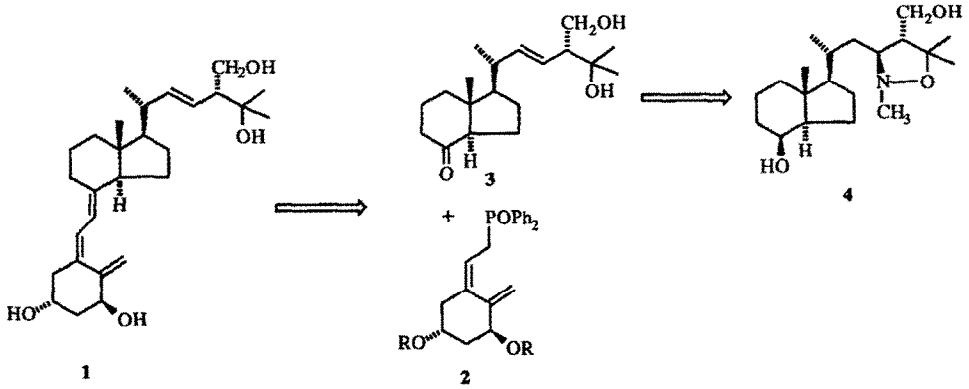
Abstract: A convergent total synthesis of 1α,25,28-trihydroxyergocalciferol (**1**), using an available A ring synthon and a CD synthon which was prepared via a thermodynamically controlled dipolar cycloaddition of methyl 3,3-dimethylacrylate and a C-23 nitrone followed by subsequent removal of the nitrogen function, is described.

In recent years the functions of vitamin D and its metabolites have been under extensive investigation. In addition to the role in the maintenance of calcium and phosphorus homeostasis and bone resorption and mineralization, widespread distribution of vitamin D receptors in many tissues indicates that this hormone plays a much wider biological role. And, indeed, recent discoveries have shown connections to cell differentiation and cell proliferation. Already there is an established therapeutic use in the treatment of psoriasis and indications of potential use in the treatment of leukemias.[2]

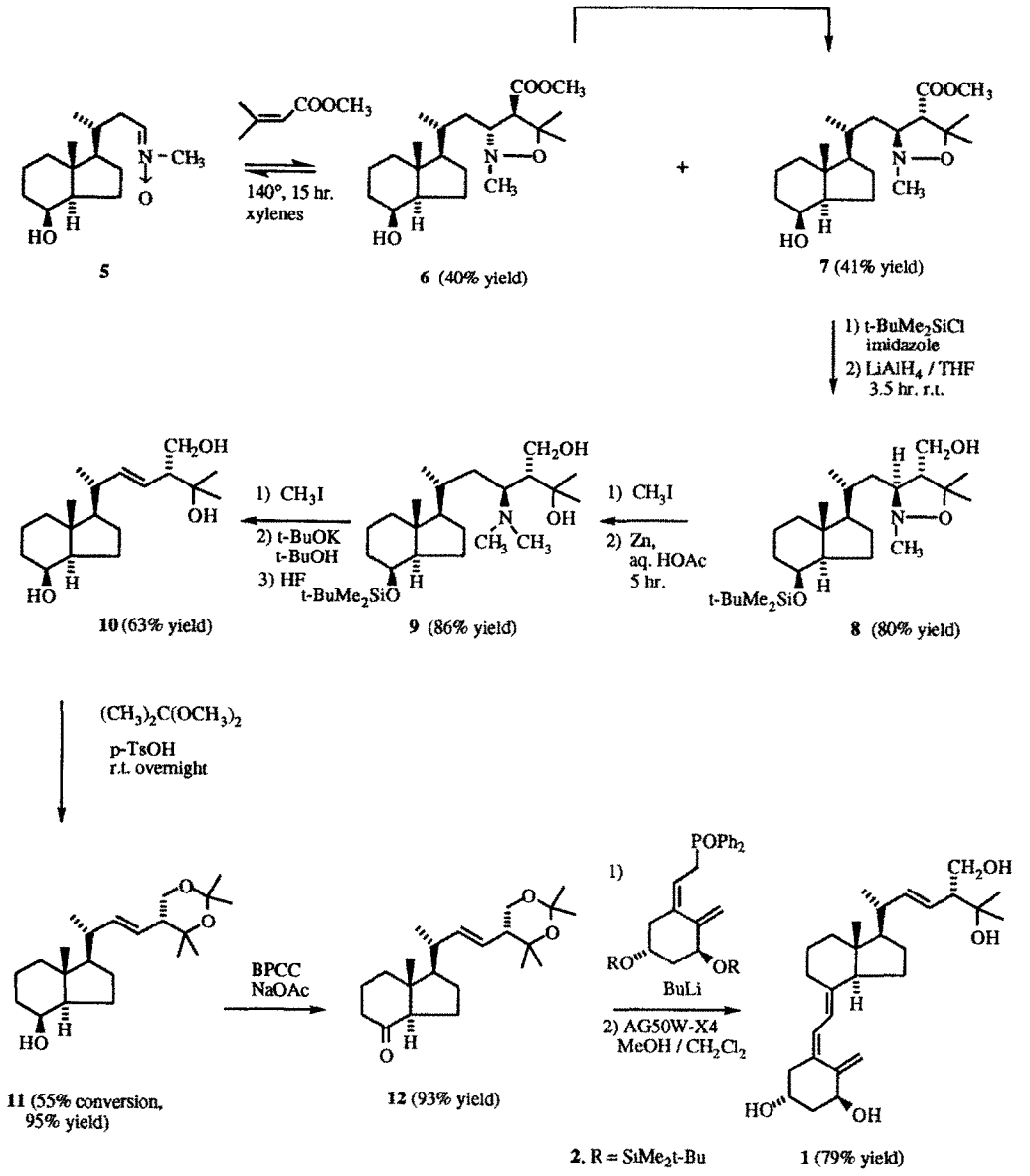
Initially we became interested in the synthesis of 1α,25,28-trihydroxyergocalciferol (**1**) because it was suspected to be the structure of a newly isolated metabolite of vitamin D_2. Although it was soon realized that this new metabolite did not have the proposed structure, 1α,25,28-trihydroxyvitamin D_2 (**1**) was nonetheless of interest in our program of making vitamin D_3 and D_2 metabolites and derivatives available for biological evaluation.

The merits of a convergent synthesis, based on the concept of Lythgoe,[3] have made this approach overwhelmingly the method of choice for the preparation of numerous vitamin D metabolites and analogs.

Scheme 1



Scheme 2


COOCH3
N - CH3
O
HO
H
5
140°, 15 hr.
xylenes
N—O
CH3
6 (40% yield)
+
7 (41% yield)
1) t-BuMe2SiCl
imidazole
2) LiAlH4 / THF
3.5 hr. r.t.
CH2OH
OH
1) CH3I
2) t-BuOK
t-BuOH
3) HF
t-BuMe2SiO
10 (63% yield)
9 (86% yield)
1) CH3I
2) Zn,
aq. HOAc
5 hr.
8 (80% yield)
(CH3)2C(OCH3)2
p-TsOH
r.t. overnight
BPCC
NaOAc
11 (55% conversion,
95% yield)
12 (93% yield)
POPh2
1)
RO
OR
BuLi
2) AG50W-X4
MeOH / CH2Cl2
2. R = SiMe2t-Bu
1 (79% yield)

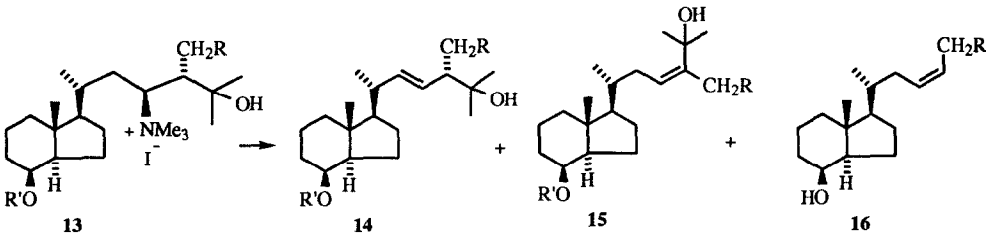
With access to the A-ring synthon (**2, Scheme 1**) already secure,[4,5] we focused our attention on the preparation of an appropriately protected CD synthon.[6] Our strategy for this synthon, exhibiting a branched nine carbon side chain with five contiguously functionalized carbon atoms, evolved from our previous experience with thermodynamically controlled nitrone-acrylate ester dipolar cycloadditions as convenient routes to vitamin D derivatives.[4,5,7,8,9] The use of acrylate esters as dipolarophiles is particularly advantageous because the regioselectivity of the cycloadditions is predictable and the cycloadditions are reversible, so even in cases where there is no diastereofacial selectivity, the undesired isomer (isomers) can be separated and re-equilibrated, thereby gaining control over the desired isomer production. In the light of our previously reported synthesis of 1α, 25-dihydroxyergocalciferol, the potential utility of the isoxazolidine intermediate **4** seemed particularly enticing.

The hydroxymethylisoxazolidine **4** (**Scheme 1**) had been obtained by lithium aluminum hydride reduction of the ester **7** (**Scheme 2**). Ester **7** and isomeric ester **6** had been produced in essentially equal amounts from a regioselective *endo* cycloaddition of the *Z*-nitrone **5**[4] and methyl 3,3-dimethylacrylate. As previously reported the undesired ester **6** had been separated by chromatography and had been re-equilibrated by heating in xylenes with an excess of dipolarophile to regenerate a 1:1 mixture of isomeric esters **6** and **7**.

On the basis of our previous experience, we prepared the unprotected hydroxymethylisoxazolidine **4** in anticipation that excision of the nitrogen function would produce **10** (**Scheme 2**) in a straightforward uneventful manner; however, this proved not to be the case. In our earlier work on the synthesis of 1α,25-$(OH)_2D_2$, the Hofmann degradation of **13a** (R=R'=H, **Table 1**), with potassium *tert*-butoxide in *tert*-butanol at reflux for 5 hours resulted in an 78% yield of the desired *trans*-Δ^{22}-olefin **14a** accompanied by 20% of olefin **16a**, the result of a Grob fragmentation. In contrast, under these same conditions with quaternary ammonium salt **13b** (R=OH, R'=H),[10] the fragmentation pathway predominated, giving rise to a 45:55 ratio of *trans*-Δ^{22}-olefin **14b** and fragmentation product **16b**, respectively.

We then discovered that the Grob fragmentation could be completely suppressed by using a stronger base allowing the elimination to be carried out at lower temperatures (LDA–THF at -5 °C); however, the regioselectivity of the Hofmann elimination was compromised by the formation of appreciable amounts of undesired Δ^{23}-olefin. Initially we carried out this elimination on the 8-*tert*-butyldimethylsilyloxy methiodide **13c** which afforded a chromatographically inseparable 2:1 (NMR) mixture of Δ^{22}- and Δ^{23}-olefins **14c** and **15c**, respectively. Methiodide **13c** was prepared from the ester **7** as seen in **Scheme 2** by 1) silylation, 2) $LiAlH_4$ reduction to **8**, 3) CH_3I, 4) Zn–HOAc reduction to **9**, and 5) quaternization with CH_3I. The mixture of Δ^{22}- and Δ^{23}-silyl ethers **14c** and **15c** was desilylated (HF–THF–CH_3CN), and the free *trans*-Δ^{22}-alcohol **14b** (=**10**) was easily obtained pure (with no contaminating Δ^{23}-alcohol **15b**) in 63% yield after chromatography. The remainder of the material eluted from the silica gel column was not Δ^{23}-alcohol **15b**, but appeared, by NMR, to be a mixture of dienes presumably from loss of the tertiary allylic hydroxyl at C-25 under the acidic conditions of the desilylation. We subsequently found that the elimination of the hydroxy quaternary salt **13b** also gave about the same Δ^{22} to Δ^{23} product ratio (NMR) as did silyl ether **13c**, but the isolated yield of Δ^{22}-alcohol **14b**, which was inseparable from Δ^{23}-alcohol **15b** on untreated silica gel, was only 47% after extensive chromatography on argentiated silica gel.[11]

Table 1. Hofmann Degradation of 13



Starting Material			Conditions	Product Ratio 14 : 15 : 16	Isolated Yield of 14
13a	R = H,	R' = H	t-BuOK–t-BuOH, reflux, 5 hr,	78 : 0 : 20	78%[4]
13b	R = OH,	R' = H	t-BuOK–t-BuOH, reflux, 5 hr,	45 : 0 : 55	34%
13c	R = OH,	R' = $SiMe_2$t-Bu	LDA–THF, -5 C, 3.5 hr	67 : 33 : 0	63%†
13b	R = OH,	R' = H	LDA–THF, -50° to 0 °C, 3.5 hr	67 : 33 : 0	47%

† Yield after desilylation (R' = OH).

The remaining steps proceeded uneventfully (**Scheme 2**). The side chain primary and tertiary alcohols of **10** were selectively protected by conversion to the hydroxyacetonide, **11.** Oxidation of **11** to protected ketone **12** proceeded in 93% yield with bipyridinium chlorochromate (BPCC). Wittig-Horner coupling of ketone **12** with the lithiated A ring synthon **2** followed by removal of the acetonide and *tert*-butyldimethylsilyl functions gave 1α,25,28-trihydroxyergocalciferol (**1**)[12] in 79% yield.

The accompanying paper in this issue discloses the enhancement of 1α,25-$(OH)_2D_3$ specific nuclear receptor expression *in vivo* without apparent hypercalcemia on treatment with 1α,25,28-$(OH)D_2$ (**1**).

NOTES AND REFERENCES

1. Deceased September 30, 1988
2. For reviews covering chemistry and biology of vitamin D derivatives see: Turner, A. B. *Nat. Prod. Rep.*, **1989**, *6*, 53. Reichel, H.; Koeffler, H. P.; Norman, A. W. *N. Engl. J. Med.*, **1989**, *320*, 980; Holick, M. F. *Arch. Dermatol.*, **1989**, *125*, 1692; Vitamin D: Gene Regulation, Structure-Function Analysis, and Clinical Application, Proc. of the Eighth Workshop on Vitamin D, de Gruyter, W. New York, **1991**; Vitamin D. Molecular, Celluar and Clinical Endocrinology, Proc. of the Seventh Workshop on Vitamin D, Norman, A. W.; Schaefer, K.; Gringoleit, H. -G.; Herrath, D. V.; de Gruyter, W. New York, **1988**.
3. (a) Lythgoe, B.; Moran, T. A.; Nambudiry, M. E. N.; Ruston, S.; Tideswell, J.; Wright, P. W. *Tetrahedron Lett.* **1978**, 3863. (b) Lythgoe, B.; Moran, T. A.; Nambudiry, M. E. N.; Ruston, S. *J. Chem. Soc., Perkin Trans. 1* **1976**, 2386. (c) Lythgoe, B.; Nambudiry, M. E. N.; Tideswell, J. *Tetrahedron Lett.* **1977**, 3685. (d) Lythgoe, B.; Manwaring, R.; Milner, J. R.; Moran, T. A.; Nambudiry, M. E. N.; Tideswell, J. *J. Chem. Soc., Perkin Trans. 1* **1978**, 387. (e) Lythgoe, B.; Moran, T. A.; Nambudiry, M. E. N.; Tideswell, J.; Wright, P. W. *J. Chem. Soc. Perkin Trans. 1* **1978**, 590 (f) Kocienski, P. J.; Lythgoe, B. *J. Chem. Soc., Perkin Trans. 1* **1979**, 1290. (g) Lythgoe, B. *Chem. Soc. Rev.* **1981**, 449.
4. (a) Baggiolini, E. G.; Iacobelli, J. A.; Hennessy, B. M.; Batcho, A. D.; Sereno, J. F.; Uskoković, M. R. *J. Org. Chem.* **1986**, *51*, 3098. (b) Batcho, A. D.; Sereno, J. F.; Baggiolini, E. G.; Hennessy, B. M.; Uskoković, M. R. *Croat. Chem. Acta*, **1986**, *58*, 757.
5. Kiegel, J.; Wovkulich, P. M.; Uskoković, M. R. *Tetrahedron. Lett.* **1991**, *43*, 6057
6. After this work was completed, an elegant alternative approach to this side chain was described by Midland, M. M.; Kwon, Y. C. *Tetrahedron Lett.* **1985**, *26*, 5017
7. Wovkulich, P. M.; Barcelos, F.; Batcho, A. D.; Sereno, J. F.; Baggiolini, E. G.; Hennessy, B. M.; Uskoković, M. R. *Tetrahedron*, **1984**, *40*, 2283.
8. Batcho, A. D.; Sereno, J. F.; Chadha, N. K.; Partridge, J. J; Baggiolini, E. G.; Uskoković, M. R. *Curr. Trends Org. Synth., Proc. Int. Conf., 4th* **1983**; 193
9. Wovkulich, P. M.; Baggiolini, E. G.; Hennessy, B. M.; Uskoković, M. R. *Heterocycles*, **1993**, *35*, 0000
10. The salt **13b** was prepared from the isoxazolidine **4** by 1) methyl iodide, 2) reduction to the acylic aminoalcohol (unprotected **9**) with zinc in 75 % aq. acetic acid (analogous to **8**→**9** in **Scheme 2**), and 3) reaction with methyl iodide.
11. Most likely the silylation of **7** was unnecessary since purification of Δ^{22}-alcohol **14b** from Δ^{23}-alcohol **15b** might be accomplished more simply if the sensitive Δ^{23}-olefin **15b** is transformed by treatment of the mixture with acid.
12. Batcho, A. D.; Horst, R. L.; Uskoković, M. R.; Napoli, J. L. **U.S. Patent 4,929,609** (May 29, 1990)