Communications to the Editor

[Chem. Pharm. Bull. 36(7)2724—2725(1988)]

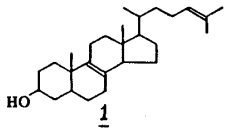
FACILE SYNTHESIS OF ZYMOSTEROL AND RELATED COMPOUNDS

Noriko Ogihara and Masuo Morisaki*
Kyoritsu College of Pharmacy, Shibakoen, Minato-ku, Tokyo 105, Japan

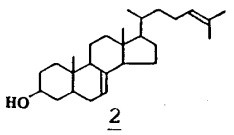
Facile preparations of cholesta-8,24-dien-3β-ol, zymosterol (1) and the related 7,24-diene (2), 5,7,24-triene (3) and 8,14,24-triene (4), all of which are potential intermediates of cholesterol or ergosterol biosynthesis, are described.

KEYWORDS —— zymosterol; cholesta-8,24-dien-3β-ol; cholesterol biosynthesis; GC-MS; cholesta-5,7,24-trien-3β-ol; cholesta-7,24-diene-3β-ol

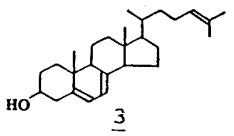
One of the key intermediates in the biosynthesis of cholesterol (in mammals) and ergosterol (in yeast and fungi) is zymosterol, cholesta-8,24-dien-3β-ol.[1] The only available standard sample of zymosterol has been from minor yeast sterols,[2] until the very recent report[3] of a chemical synthesis of zymosterol, which has prompted us to report our own efforts in this area. Described here is a facile way to prepare zymosterol (1), cholesta-7,24-dien-3β-ol (2), cholesta-5,7,24-trien-3β-ol (3) and cholesta-8,14,24-trien-3β-ol (4), all of which are potential precursors of cholesterol or ergosterol.



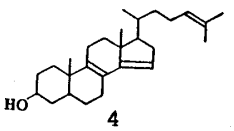
1



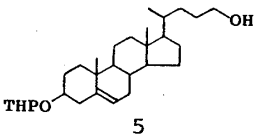
2



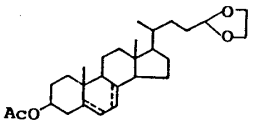
3



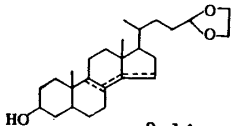
4



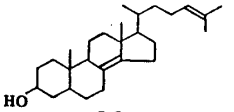
5



6, Δ^5
7, $\Delta^{5,7}$
8, Δ^7



9, $\Delta^{8,14}$
10, $\Delta^{8(9)}$
11, $\Delta^{8(14)}$



12

5-Cholene-3β,24-diol 3-tetrahydropyranyl ether(5)[4] is oxidized with pyridinium chlorochromate to give the 24-aldehyde, which is treated with ethylene glycol in refluxing benzene in the presence of p-toluenesulfonic acid. The resultant 3-hydroxy-24-acetal is converted into the acetate(6), mp 124-126°C in 75% overall yield from 5. Successive treatment of 6 with N-bromosuccinimide in refluxing carbon tetrachloride, tetra-n-butylammonium bromide in tetrahydrofuran, and tetra-n-butylammonium fluoride[5] gives the 5,7-diene (7, 43% yield), the common synthetic progenitor of all the targeted sterols. Catalytic hydrogenation of the 5,7-diene (7) with Raney-Ni W4 in ethanol affords the 7-ene (8). Refluxing 7 with benzene in the presence of p-toluenesulfonic acid affords the 8,14-diene (9) in 55% yield, after basic hydrolysis. Similar catalytic hydrogenation of

the 8,14-dien-3β-ol(**9**) gives a mixture (ca.1:1) of the 8(9)-ene (**10**, ^{1}H-NMR 0.61 ppm, 13-Me) and the 8(14)-ene (**11**, 0.69 ppm).[6]

Deacetalization of the corresponding acetates of **10** plus **11** with d.HCl in acetone followed by Wittig reaction with isopropylidene triphenylphosphorane in tetrahydrofuran yields, after saponification, a mixture (ca.1:1) of zymosterol(**1**) and the 8(14),24-diene (**12**) in 40% yield. Recrystallization of this material from methanol gave zymosterol, mp 109-111°C (lit.[2a,3] mp 110-112°C). In the same manner, deacetalization of **7**, **8**, and **9** followed by Wittig reaction gives the 5,7,24-triene (**3**), 7,24-diene (**2**) and 8,14,24-triene (**4**), respectively.

These sterols in biological systems are identified by HPLC and/or GC-MS. The relevant data to use for that purpose are listed in Table I. A characteristic fragment ion of Δ^{24} steroid trimethylsilyl (TMS) ether is m/z 343 due to the loss of the side chain together with two hydrogen atoms from the steroid nucleus,[7] as exemplified by the mass spectra of the 7,24-diene(**2**) and desmosterol TMS ethers. However, in the mass spectra of the TMS ethers of **1**, **3** and **4**, the corresponding peak (m/z,341 in **3** and **4**) almost completely disappears. Thus, fragmentations induced by the 24-double bond are highly dependent on the position of the nuclear double bond.

Table I

	HPLC[a] t_R(min)	GC[b] RRT	Prominent mass fragment ions(%)[c]
Cholesterol	15.8 min	1.00	458(42), 368(75), 329(85), 129(100)
Desmosterol	13.6	1.09	456(28), 366(26), 343(64), 69(100)
Zymosterol(**1**)	12.2	1.13	456(100), 441(42), 351(28)
7,24-Diene(**2**)	13.2	1.25	456(33), 441(30), 343(100)
8(14),24-Diene(**12**)	11.7	1.10	456(45), 441(26), 343(44), 69(100)
5,7,24-Triene(**3**)	11.2	1.19	454(69), 364(32), 349(100), 323(68)
8,14,24-Triene(**4**)	9.8	1.10	454(100), 439(29), 369(27), 349(31)

a) Shim-pack CLC-ODS, 0.15m x 6.0 ; methanol, 1.0 ml/min; UV detector, 210 nm.
b) As the TMS ether; capillary column, Ultra 1(methyl silicon); 260°C.
c) As the TMS ether; 70 eV.

REFERENCES AND NOTES

1) G. J. Schroepfer, Jr., Annu. Rev. Biochem., **51**, 555(1982).
2) a) U. F. Taylor, A. Kisic, R. A. Pascal, Jr., A. Izumi, M. Tsuda and G. J. Schroepfer, Jr., J. Lipid Res., **22**, 171(1981); b) N. Ariga, H. Hatanaka, J. Nagai and H. Katsuki, J. Biochem., **83**, 1109(1978).
3) R. E. Dolle, S. J. Schmidt and L. I. Kruse, J. Chem. Soc., Chem. Commun., **1988**, 19.
4) M. Morisaki, M. Shibata, C. Duque, N. Imamura and N. Ikekawa, Chem. Pharm. Bull., **28**, 606(1980).
5) M. P. Rappapold, J. Hoogendorf and L. F. Pauli, "Vitamin D. Chemical Biological and Clinical Endocrinology of Calcium Metabolism," Walter de Gruyter and Co., Berlin, 1982, p. 1133.
6) Catalytic hydrogenation of the 8,14-dienic steroids over Raney-Ni was reported to give the 8(14)-olefin,[a] 8(9)-olefin,[b] or their mixture.[c] a) M. A. Apfel, J. Org. Chem., **44**, 643(1979); b) P. J. Hylands, J. M. Midgley, C. Smith, A. F. A. Wallis and W. B. Whalley, J. Chem. Soc., Perkin Trans. 1, **1977**, 817; c) D. H. R. Barton and J. D. Cox, J. Chem. Soc., **1949**, 214.
7) S. G. Wyllie and C. Djerassi, J. Org. Chem., **33**, 305(1968); I. J. Massey and C. Djerassi, J. Org. Chem., **44**, 2448(1979).

(Received May 2, 1988)