

Synthesis of 24-Methylene-cholest-5-ene-7-oxo-3 β ,19-diacetate

Wei Gang LU, Cui Xian ZHANG, Jing Yu SU, Long Mei ZENG*

School of Chemistry and Chemical Engineering, Sun Yat-Sen University, Guangzhou 510275

Abstract: 24-Methylenecholest-5-ene-7-oxo-3 β ,19-diacetate **5** was synthesized starting from stigmasterol *via* twelve step reactions in 4.1% overall yield. It can be served as a key intermediate for the synthesis of 24-methylenecholest-5-ene-3 β ,7 β ,19-triol **1**, a naturally occurring polyhydroxylated sterol with potential biological activity.

Keywords: 24-Methylene-cholest-5-ene-3 β ,7 β ,19-triol, synthesis, allylic oxidation, PCC/Al₂O₃.

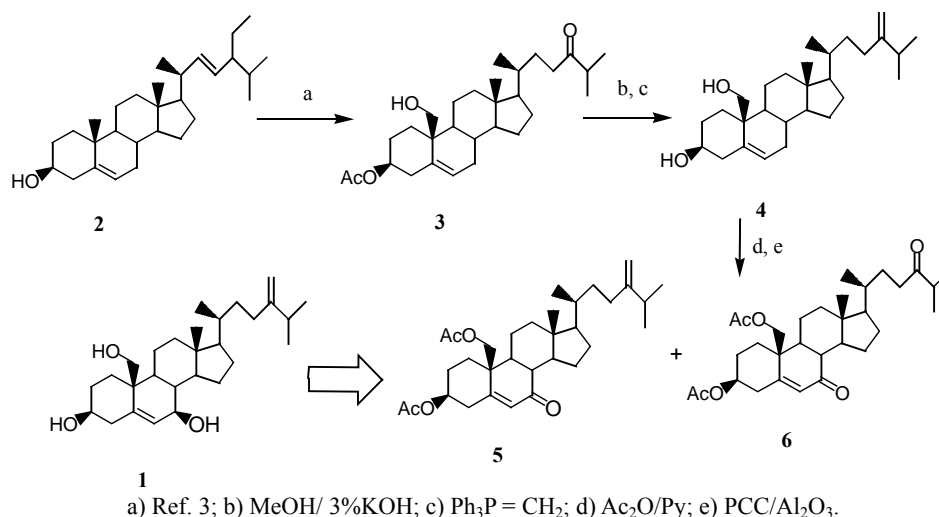
24-Methylenecholest-5-ene-3 β , 7 β , 19-triol **1**, isolated from the soft corals *Nephthea albida* and *Nephtea tiexieral verseveldt* by L. M. Zeng¹, exhibited significant cytotoxicity against A549 (human lung adenocarcinoma), H-29 (human colon adenocarcinoma), KB (human epidemoid carcinoma), and P-388 (murine lymphocytic leukermia) cell lines with ED₅₀ values of 0.69, 0.72, 0.58, and 0.24 μ g/mL, respectively². This spurred our interest in the synthesis of compound **1**. In the designed synthetic route (as shown in **Scheme 1**), 24-methylenecholest-5-ene-7-oxo-3 β ,19-diacetate **5** is a key intermediate, which can be easily transformed to **1** with the stereospecific reduction of 7-carbonyl group and saponification of C-3 acetate. Herein, we report the synthesis of **5**.

Conversion of stigmasterol **2** to **3** was accomplished in 8 steps referring to the literature³ in 24% overall yield. Compound **3** was transformed to **4** with hydrolysis and Wittig reaction⁴ in 59% yield. After protection of 3 β , 19-dihydroxy groups with Ac₂O/pyridine, allylic oxidation⁵ of **4** was carried out using PCC/Al₂O₃ (pyridinium chlorochromate absorbed on alumina)⁶ in the presence of anhydrous benzene to yield the expected product, 24-methylenecholest-5-ene-7-oxo-3 β ,19-diacetate **5** and the unexpected product, cholest-5-ene-7,24-dioxo-3 β ,19-diacetate **6**. Both **5** and **6** are new compounds (**Scheme 1**).

PCC/Al₂O₃ had been first applied in allylic oxidation of steroids with 5,6-olefin, and it turned out to be effective, moreover, oxidation with PCC/Al₂O₃ was profitable in the working up, which was reduced to a mere filtration. The amount of PCC/Al₂O₃ was important in this reaction; too much PCC/Al₂O₃ was favorable for the formation of **6**. A reasonable reaction condition was obtained after extensive investigation, **5** and **6** were obtained from **4** in 29%, 14% yield, respectively (**Table 1**).

* E-mail: ceszlm@zsu.edu.cn

Scheme 1

**Table 1** Oxidation of 24-methylenecholest-5-ene-3 β ,19-diacetate with PCC/Al₂O₃*

Ratio of oxidant/substrate (g / g)	Recovered substrate (%)	5 (%**)	6 (%**)
20	45	11	6
30	34	18	8
40	20	29	14
50	21	26	14
60	16	25	17
80	12	21	20

* The reaction was carried out in refluxing anhydrous benzene under N₂ for 24 hrs.

** The yield was calculated after separation by column chromatography.

** The yield was calculated without the concern of the recovered substrate.

This synthetic route seemed to be feasible, and it was better than when pursuing an alternative route, which involved the protection of the C-24 carbonyl group as a ketal, allylic oxidation at C-7, and then stereoselective reduction of the resulting carbonyl group with NaBH₄/CeCl₃, followed by removal of the ketal and saponification of the C-3 acetate⁷. Pursuing methylenation before introduction of 7 β -hydroxy group not only saved the procedures of protection and deprotection of the C-24 carbonyl group, but also had reasonable yield obtained.

The IR spectrum of **5** showed medium C–H stretching at 3080 cm⁻¹, C=C stretching at 1640 cm⁻¹ and C–H out-of-plane stretching at 887 cm⁻¹, corresponding to the 24(28) terminal double bond, strong stretching at 1674 cm⁻¹ was assigned to α,β -unsaturated ketone; its ¹H NMR spectrum showed two doublets at δ 4.66 (brs) and 4.72 (brs) for 28-H₂; the signals at δ 201.6, 156.8, 106.3 in the ¹³C NMR spectrum of **5** verified the existence of α,β -unsaturated ketone and terminal double bond⁸. The IR spectrum of **6** showed saturated ketone stretching at 1711cm⁻¹, and no characteristic adsorption of terminal double bond; its ¹H NMR and ¹³C NMR spectra also showed the absence of

terminal double bond, and the signals at δ 215.4, 201.6 in the ^{13}C NMR spectrum of **6** verified the existence of saturated ketone and α,β -unsaturated ketone⁹.

Acknowledgments

We are grateful to the National Natural Science Foundation of China (No. 29932030), the National High Technology Development Project (No. 2001AA624030), and the Natural Science Foundation of Guangdong Province (No. 04300569) for financial support for this work.

References and Notes

1. G. Y. S. Wang, F. Y. Li, L. M. Zeng, *Chem. J. Chin. Univ.*, **1992**, *13*, 623.
2. C. Y. Duh, S. K. Wang, M. J. Chu, *J. Nat. Prod.*, **1998**, *61*, 1022.
3. W. G. Lu, M. Y. Wang, J. Y. Su, L. M. Zeng, *Chin. Chem. Lett.*, **2002**, *13*, 119.
4. W. G. Lu, J. Y. Su, L. M. Zeng, *Chem. J. Chin. Univ.*, **2004**, *25*, 1648.
5. E. J. Parish, T. Y. Wei, *Synthetic Communications*, **1987**, *17*, 1227.
6. Y. S. Cheng, W. L. Liu, S. H. Chen, *Synthesis*, **1980**, *3*, 223.
7. C. E. Heltzel, A. A. L. Gunatilaka, D. G. I. Kingston, *et al.*, *J. Nat. Prod.*, **1994**, *57*, 620.
8. Physical data of compound **5**: Colorless viscous solid; IR (KBr, cm^{-1}) 3081, 2956, 2870, 1741, 1674, 1640, 1464, 1445, 1369, 1232, 1038, 887; ^1H -NMR (CDCl_3 , δ ppm) 0.71 (s, 3H, H-18), 0.96 (d, 3H, $J = 6.5\text{Hz}$, H-21), 1.02 (d, 3H, $J = 7.0\text{Hz}$, H-26 or H-27), 1.03 (d, 3H, $J = 7.0\text{Hz}$, H-26 or H-27), 2.03 (s, 3H, 3-OCOCH₃ or 19-OCOCH₃), 2.05 (s, 3H, 3-OCOCH₃ or 19-OCOCH₃), 4.12 (d, 1H, $J = 12\text{Hz}$, Ha-19), 4.66 (brs, 1H, Ha-28), 4.71 (d, 1H, $J = 12\text{Hz}$, Hb-19), 4.72 (brs, 1H, Hb-28), 4.74 (m, 1H, H-3), 5.89 (d, 1H, $J = 1.5\text{Hz}$, H-6); ^{13}C -NMR (CDCl_3 , δ ppm) 201.6, 170.5, 170.4, 158.3, 156.8, 130.0, 106.3, 72.1, 64.6, 55.0, 51.5, 50.2, 47.0, 43.8, 41.8, 39.4, 38.3, 36.0, 35.0, 34.1, 33.5, 31.4, 30.1, 28.0, 26.5, 22.4, 22.2, 22.1, 21.6, 21.3, 19.2, 12.5.
9. Physical data of compound **6**: Colorless viscous solid; IR (KBr, cm^{-1}) 2929, 2863, 1738, 1711, 1665, 1464, 1445, 1369, 1236, 1036; ^1H -NMR (CDCl_3 , δ ppm) 0.70 (s, 3H, H-18), 0.92 (d, 3H, $J = 6.5\text{Hz}$, H-21), 1.09 (d, 3H, $J = 7.0\text{Hz}$, H-26 or H-27), 1.09 (d, 3H, $J = 3.0\text{Hz}$, H-26 or H-27), 2.03 (s, 3H, 3-OCOCH₃ or 19-OCOCH₃), 2.05 (s, 3H, 3-OCOCH₃ or 19-OCOCH₃), 4.12 (d, 1H, $J = 12\text{Hz}$, Ha-19), 4.71 (d, 1H, $J = 12\text{Hz}$, Hb-19), 4.74 (m, 1H, 3-H), 5.89 (brs, 1H, 6-H); ^{13}C -NMR (CDCl_3 , δ ppm) 215.4, 201.6, 170.5, 170.4, 158.3, 130.0, 72.1, 64.7, 54.9, 51.4, 50.2, 47.0, 43.8, 41.8, 41.2, 39.3, 38.3, 37.6, 35.6, 33.5, 30.2, 30.1, 28.0, 26.5, 22.0, 21.6, 21.3, 19.0, 18.8, 18.7, 12.5.

Received 8 June, 2004