Chemistry and Physics of Lipids 163 (2010) 165–171

Contents lists available at ScienceDirect

Chemistry and Physics of Lipids

journal homepage: www.elsevier.com/locate/chemphyslip

Oxyfunctionalization of unactivated C–H bonds in triterpenoids with *tert*-butylhydroperoxide catalyzed by *meso*-5,10,15,20-tetramesitylporphyrinate osmium(II) carbonyl complex

Shoujiro Ogawa [a], Yasuo Wakatsuki [a], Mitsuko Makino [b], Yasuo Fujimoto [b], Ken Yasukawa [c], Takashi Kikuchi [d], Motohiko Ukiya [d], Toshihiro Akihisa [d], Takashi Iida [a,*]

[a] *Department of Chemistry, College of Humanities and Sciences, Nihon University, Sakurajousui, Setagaya-ku, Tokyo 156-8550, Japan*
[b] *Department of General Studies, College of Humanities and Sciences, Nihon University, Sakurajousui, Setagaya-ku, Tokyo 156-8550, Japan*
[c] *College of Pharmacy, Nihon University, 7-7-1 Narashinodai, Funabashi, Chiba 274-8555, Japan*
[d] *Department of Materials and Applied Chemistry, College of Science and Technology, Nihon University, 1-8 Kanda Surugadai, Chiyoda-ku, Tokyo 101-8301, Japan*

ARTICLE INFO

Article history:
Received 24 August 2009
Received in revised form 29 October 2009
Accepted 29 October 2009
Available online 10 November 2009

Keywords:
Allylic oxidation
Oxyfunctionalization
Cytochrome P-450
Hydroxylation
Ketonization
Triterpenoid
Unactivated C–H bond

ABSTRACT

A system consisting of *meso*-5,10,15,20-tetramesitylporphyrinate osmium(II) carbonyl complex [Os(TMP)CO] as a precatalyst and *tert*-butylhydroperoxide (TBHP) as an oxygen donor is shown to be an efficient, regioselective oxidant system for the allylic oxidation, ketonization and hydroxylation of unactivated C–H bonds in a series of the peracetate derivatives of penta- and tetracyclic triterpenoids. Treatment of the substrates with this oxidant system afforded a variety of novel or scarce oxygenated derivatives in one-step. Structures of the isolated components, after chromatographic separation, were determined by spectroscopic methods including GC–MS and shift-correlated 2D-NMR techniques. Factors governing the regioselectivity and the possible mechanism for the oxyfunctionalization of the unactivated carbons are also discussed.

1. Introduction

In analogy with enzyme-controlled reactions *in vivo*, such as cytochrome P-450 oxidase-dependent systems (Sono et al., 1996; Makris et al., 2006), the oxyfunctionalization of unactivated carbon atoms is a key transformation in the efficient and convenient synthesis of biologically active compounds, starting from abundantly available, naturally occurring compounds such as steroids, alkaloids, and bile acids. In particular, the combination of the cytochrome P-450 analogs, metalloporphyrin precatalyst/oxygen-donar systems (Breslow, 1990; Yang et al., 2000, 2002; Reese, 2001; Meunier, 1992; Breslow et al., 1997; Ohtake et al., 1995; Shingaki et al., 1997) and the smallest three-membered cyclic peroxide, dioxiranes (Reese, 2001; Bovicelli et al., 1992; Iida et al., 2001), have been shown to insert efficiently an oxygen atom into unactivated methine and/or methylene C–H bonds in these substrates.

We have recently developed a novel, powerful oxidant system, which consists of *meso*-5,10,15,20-tetramesitylporphyrinate osmium (II) carbonyl complex [Os(TMP)CO] as a precatalyst and *tert*-butylhydroperoxide (TBHP) as an oxygen donor (Ogawa et al., 2007a; Iida et al., 2007). The oxidant system proceeds through an active *trans*-dioxoosmiumporphyrin intermediate with a high turnover rate (*ca.* 200:1) as a catalyst and oxidizes regioselectively unactivated tertiary methine CH and/or secondary methylene CH_2 protons to yield the corresponding hydroxy- and oxo-derivatives, respectively. As a part of our ongoing program to produce bioactive compounds from available natural products, we became interest in the useful, efficient application of the Os(TMP)CO/TBHP system to terpenoids. We describe here the regioselective oxyfunctionalization of a series of penta- and tetracyclic triterpenoids using this oxidant system. An additional aim of the study was to prepare novel or scarece oxygenated derivatives of potential medicinal significance.

2. Experimental

2.1. General

Melting points (mp) were determined on an electric micro-hot stage and are uncorrected. IR spectra were obtained on a

* Corresponding author. Tel.: +81 3 3329 1151x5704; fax: +81 3 3303 9899.
E-mail address: takaiida@chs.nihon-u.ac.jp (T. Iida).

0009-3084/$ – see front matter © 2009 Elsevier Ireland Ltd. All rights reserved.
doi:10.1016/j.chemphyslip.2009.10.012

JASCO FT-IR 4100 spectrometer (Tokyo, Japan) for samples in the KBr tablets. ^{1}H and ^{13}C NMR spectra were measured on a JEOL JNM-EX270 FT instrument (Tokyo, Japan) in $CDCl_3$ containing 0.1% Me_4Si as an internal standard. Chemical shifts are expressed as δ ppm relative to Me_4Si and coupling constants as in Hz. ^{13}C NMR signals corresponding to methyl (CH_3), methylene (CH_2), methine (CH), and quaternary (C) carbons were differentiated by means of distortionless enhancement by polarization transfer (DEPT) experiments. 2D-NMR spectra were measured on a JEOL GSX-400 spectrometer or Verian Mercury 300BB (CA, USA). Low-resolution electron-impact mass (LR-EI-MS) spectra were recorded on a JEOL JMS-GCmate mass spectrometer at 70 eV using the positive ion mode (PIM). High-resolution electron-impact mass spectra (HR-EI-MS) were measured on a JEOL GCmate mass spectrometer. A Shimadzu GC-2010 gas chromatograph equipped with a flame ionization detector was used isothermally at 320 °C fitted with a chemically bonded, fused silica capillary column (25QCC3/BPX5; 25 m × 0.32 mm i.d.; film thickness, 0.25 μm; SGE, Melbourne, Australia). The preparative HPLC apparatus consisted of a Hitachi (Tokyo, Japan) L-7100 pump equipped with a Shodex RI detector (Tokyo, Japan) and a Pegasil Silica 60-5 column (250 mm × 10 mm i.d., Tokyo, Japan); hexane-EtOAc mixtures (9:1–7:3,v/v) were used as the eluent. Thin-layer chromatography (TLC) was performed on precoated silica gel plates (0.25 mm layer thickness; E. Merck, Darmstadt, Germany) using hexane-EtOAc mixtures as the developing solvent. 70% TBHP was purchased from Tokyo Kasei Kogyo Co. (Tokyo, Japan); it was extracted with CH_2Cl_2 and the organic layer was evaporated under reduced pressure prior to use.

2.2. Materials and reagents

meso-Tetraarylporphyrins were prepared by a slight modification of the procedure of Lindsey et al. (1987). The Os(TMP)CO complex was prepared from the *meso*-tetramesitylporphyrins and $Os_3(CO)_{12}$ by the method reported by Che et al. (1985). α-Amyrin, faradiol, betulin, cycloartenol, and euphol were from our laboratory collection. Acetylation of the hydroxyl groups in these compounds was carried out by the usual manner. Subsequent catalytic hydrogenation of the resulting acetate derivatives with Pd/C catalyst afforded the starting compounds (**1**–**5**) used in this study, except for **1** and **5**.

2.3. General oxidation procedure with Os(TMP)CO/TBHP

To a solution of substrate (0.2 mmol) in benzene (500 μl), molecular sieves (50 mg; 4 Å), Os(TMP)CO (1 mg, 1 μmol) and anhydrous TBHP (360 mg, 4.0 mmol) were added, and the mixture was refluxed for 48–96 h; the reaction was monitored by TLC; 48 h for **1**, 96 h for **2**-**4**, and 72 h for **5**. After the reaction, each of the oxidation products was isolated by a column chromatography on silica gel, followed by preparative HPLC.

2.4. Physicochemical data for oxidation products

11-Oxo-urs-12-en-3β-yl acetate (6). Colorless amorphous solid, isolated from the oxidation product of **1**, which was crystallized from aqueous methanol, mp 279–281 °C (lit. (Dasgupta et al., 1980), mp 276–278 °C); IR (KBr) cm^{-1} 1655, 1734 (C=O); ^{1}H NMR ($CDCl_3$, 270 MHz) δ 0.80 (3H, d, J = 5.7 Hz, H-29), 0.82 (3H, s, H-23), 0.88 (3H, s, H-24), 0.88 (3H, d, J = 1.2 Hz, H-30), 0.95 (3H, s, H-28), 1.04 (3H, s, H-25), 1.17 (3H, s, H-26) 1.29 (3H, s, H-27), 2.05 (3H, s, CH_3CO), 4.52 (1H, dd, J = 5.4, 11.6 Hz, H-3α), 5.54 (1H, s, H-12); ^{13}C NMR ($CDCl_3$, 67.5 MHz) δ 38.9 (C-1), 23.6 (C-2), 80.6 (C-3), 38.0 (C-4), 55.0 (C-5), 18.5 (C-6), 32.8 (C-7), 40.9 (C-8), 61.4 (C-9), 36.8 (C-10), 199.7 (C-11), 130.4 (C-12), 165.0 (C-13), 43.6 (C-14), 28.8 (C-15), 27.2 (C-16), 45.1 (C-17), 59.0 (C-18), 39.2 (C-19), 39.3 (C-20), 33.9 (C-21), 30.9 (C-22), 28.0 (C-23) 16.7 (C-24), 16.5 (C-25), 20.5 (C-26), 27.5 (C-27), 27.5 (C-28), 17.4 (C-29), 21.3 (C-30), 21.1 ($\underline{CH_3}CO$), 171.0 ($CH_3\underline{CO}$); LR-EI-MS (PIM) m/z 482 (M^+, 18), 422 (M-CH_3COOH, 7), 407 (M-CH_3COOH-CH_3 4), 273 (100), 232 (63).

20α-Hydroxy-ursan-3β,16β-diyl diacetate (7). Colorless amorphous solid, isolated from the oxidation product of **2**, which was crystallized from acetone, mp 222–224 °C; IR (KBr) cm^{-1} 3492 (OH), 1726 (C=O); ^{1}H NMR ($CDCl_3$, 400 MHz) δ 0.83 (3H, s, H-24), 0.85 (3H, s, H-23), 0.87 (3H, s, H-25), 0.90 (3H, s, H-28), 1.05 (3H, s, H-26), 1.06 (3H, s, H-27), 1.08 (3H, d, J = 4.6 Hz, H-29), 1.18 (3H, s, H-30), 2.00 and 2.04 (each 3H, s, CH_3CO), 4.47 (1H, dd, J = 4.2, 11.2 Hz, H-3α), 4.75 (1H, dd, J = 3.8, 11.5 Hz, H-16α); ^{13}C NMR ($CDCl_3$, 100 MHz) δ 38.4 (C-1), 23.7 (C-2), 80.9 (C-3), 36.8 (C-4), 55.2 (C-5), 18.1 (C-6), 34.4 (C-7), 41.5 (C-8), 49.1 (C-9), 37.8 (C-10) 21.6 (C-11), 29.1 (C-12), 38.6 (C-13), 42.7 (C-14), 32.8 (C-15), 79.4 (C-16), 39.5 (C-17), 46.1 (C-18), 38.1 (C-19), 72.7 (C-20), 34.9 (C-21), 32.2 (C-22), 27.9 (C-23), 16.5 (C-24), 16.3 (C-25), 16.1 (C-26), 16.3 (C-27), 13.6 (C-28), 17.8 (C-29), 30.2 (C-30), 21.3 ($\underline{CH_3}CO$), 170.6 and 171.0 ($CH_3\underline{CO}$); LR-EI-MS (PIM) m/z 484 (M-CH_3COOH, 53), 466 (M-CH_3COOH-H_2O, 69), 424 (M-2CH_3COOH, 15), 406 (M-2CH_3COOH-H_2O, 15), 359 (30), 299 (65), 189 (100); HR-EI-MS (PIM) m/z Calc. for $C_{34}H_{56}O_5$ $[M]^+$ 544.4128, Found m/z 544.4123.

21-Oxo-lupan-3β,28-diyl diacetate (8). Colorless amorphous solid, isolated from the oxidation product of **3**, which was crystallized from chloroform-methanol, mp 251–253 °C (lit., (Sejabel et al., 1997) mp 250–254 °C); IR (KBr) cm^{-1} 1725, 1736, 1750 (C=O); ^{1}H NMR ($CDCl_3$, 300 MHz) δ 0.81 (3H, d, J = 6.9 Hz, H-29), 0.85 (3H, s, H-24), 0.86 (3H, s, H-23), 0.87 (3H, s, H-25), 1.04 (3H, s, H-27), 1.08 (3H, s, H-26), 1.15 (3H, d, J = 7.2 Hz, H-30), 1.80 (1H, d, J = 14.7 Hz, H-22α), 2.02 and 2.05 (each 3H, s, CH_3CO), 2.36 (1H, d, J = 16.8 Hz, H-22β), 3.72 (1H, d, J = 11.4 Hz, H-28), 4.37 (1H, dd, J = 1.2, 12.6 Hz, H-28), 4.47 (1H, dd, J = 6.6, 10.5 Hz, H-3α); ^{13}C NMR ($CDCl_3$, 75 MHz) δ 38.5 (C-1), 23.8 (C-2), 80.8 (C-3), 37.9 (C-4), 55.4 (C-5), 18.3 (C-6), 34.0 (C-7), 41.1 (C-8), 50.0 (C-9), 37.1 (C-10), 20.9 (C-11), 27.0 (C-12), 36.6 (C-13), 42.8 (C-14), 27.2 (C-15), 30.4 (C-16), 41.7 (C-17), 46.9 (C-18), 54.2 (C-19), 30.0 (C-20), 218.1 (C-21), 52.3 (C-22), 28.1 (C-23), 16.5 (C-24), 16.2 (C-25), 16.3 (C-26), 14.9 (C-27), 64.4 (C-28), 22.4 (C-29), 16.7 (C-30), 21.1 and 22.5 ($\underline{CH_3}CO$), 170.7 ($CH_3\underline{CO}$); LR-EI-MS (PIM) m/z 542 (M^+, 7), 482 (M-CH_3COOH, 76), 467 (M-CH_3-CH_3COOH, 25), 439 (M-2CH_3-CH_2OCOCH_3, 34), 379 (M-2CH_3-CH_3COOH-CH_2OCOCH_3, 4), 189 (100).

16-Oxo-lupan-3β,28-diyl diacetate (9). Colorless amorphous solid, isolated from the oxidation product of **3**, which was crystallized from chloroform-methanol, mp 226–228 °C; IR (KBr) cm^{-1} 1699, 1740 (C=O); ^{1}H NMR ($CDCl_3$, 300 MHz) δ 0.74 (3H, d, J = 6.3 Hz, H-29), 0.85 (3H, s, H-24), 0.86 (3H, s, H-23), 0.86 (3H, d, J = 6.6 Hz, H-30), 0.88 (3H, s, H-25), 0.90 (3H, s, H-27), 1.14 (3H, s, H-26), 1.87 (1H, d, J = 13.5 Hz, H-15α), 2.02 and 2.04 (each 3H, s, CH_3CO), 2.77 (1H, d, J = 13.8, H-15β), 4.11 (1H, d, J = 11.4 Hz, H-28), 4.45 (1H, dd, J = 5.7, 10.5 Hz, H-3α), 4.58 (1H, d, J = 11.4 Hz, H-28); ^{13}C NMR ($CDCl_3$, 75 MHz) δ 38.5 (C-1), 23.8 (C-2), 80.7 (C-3), 37.9 (C-4), 55.2 (C-5), 18.2 (C-6), 34.2 (C-7), 41.3 (C-8), 50.0 (C-9), 37.1 (C-10), 21.5 (C-11), 26.4 (C-12), 37.3 (C-13), 48.7 (C-14), 45.2 (C-15), 212.2 (C-16), 61.1 (C-17), 49.5 (C-18), 44.9 (C-19), 29.2 (C-20), 20.7 (C-21), 26.8 (C-22), 28.1 (C-23), 16.7 (C-24), 16.2 (C-25), 16.7 (C-26), 15.6 (C-27), 64.3 (C-28), 14.8 (C-29), 22.8 (C-30), 21.0 ($\underline{CH_3}CO$), 170.6 and 170.7 ($CH_3\underline{CO}$); LR-EI-MS (PIM) m/z 542 (M^+, 18), 482 (M-CH_3COOH, 79), 467 (M-CH_3-CH_3COOH, 22), 439 (M-2CH_3-CH_2OCOCH_3, 26), 379 (M-2CH_3-CH_3COOH-CH_2OCOCH_3, 12), 277 (22), 189 (100); HR-EI-MS (PIM) m/z Calc. for $C_{34}H_{54}O_5$ $[M]^+$ 542.3971, Found m/z 542.3975.

22-Oxo-lupan-3β,28-diyl diacetate (10). Colorless amorphous solid, isolated from the oxidation product of **3**, which was crystallized from chloroform-methanol, mp 228–230 °C; IR (KBr) cm^{-1} 1728, 1741 (C=O); ^{1}H NMR ($CDCl_3$, 300 MHz) δ 0.76 (3H, d, J = 6.9 Hz, H-29), 0.84 (3H, s, H-24), 0.85 (3H, s, H-23), 0.86 (3H, d, J = 6.6 Hz,