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Unusual Ozonolysis of Cholesteryl and Androst-5-en-3β-yl Acetates in the Presence of Primary Amines. Formation of A-nor-3(5→6α)abeo-6β(H)-5-oxo-steroids, Confirmed by X-ray Analysis

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The reaction of cholesteryl acetate with ozone in the presence of amines has been studied. Besides the expected cleavage products, B-seco-ketoaldehyde and its aldol condensation product, 5-hydroxy-6-formyl-B-nor-5β-cholestane derivative, the abnormal product, 3β-acetoxy-A-nor-3(5→6α)abeo-6β(H)-cholestan-5-one is formed as a result of a new, anomalous cleavage of the primary ozonide. The reaction conditions and isolation procedure were optimized for achieving good yield of the abeo-compound. The X-ray crystal structure analysis confirmed the formation of the unusual steroidal skeleton. It appears to be the first crystallographic analysis of the transformed steroid with A-nor-3(5→6α)abeo skeleton. No product resulting from the amine nucleophilic participation in the cleavage of ozonides was observed.

Key words: anomalous ozonolysis, steroids, X-ray analysis

The reaction of electrophilic ozone with nucleophilic amines has been extensively studied [1]. Amine oxide formation and side chain oxidation are the two major processes in the reaction of ozone with tertiary amines. In the case of secondary amines, the nitroxide pathway or side chain oxidation is predominant, depending on the nature of alkyl groups on nitrogen. Primary amines react with ozone to give the corresponding nitroalkanes as principal products [1].

In ozonolyses of olefins, ammonia and primary amines behave as participating solvents [2a] and the term „amozonolysis" has been introduced [3]. Secondary amines react with ozonides derived from mono- and 1,1-disubstituted olefins [4] to afford the reductive amination products, tertiary amines, in high yield. In the formation of oxaziridines as products of olefin ozonolysis in the presence of primary amines [2a,5] the intermediate α-hydroperoxyamines were detected. Recently, the role of tertiary amines in cleavage of ozonides has been studied [6] and triethylamine was found the superior reagent for decomposition of ozonides derived from variety of alkenes [4]. Ozonolysis of cholesterol received much attention [7] since 1905, due to the importance of oxysterols, which are compounds of biochemical and biomedical interest [8]. In ozonolyses of cholesterol and other Δ^5-unsaturated steroids carried out

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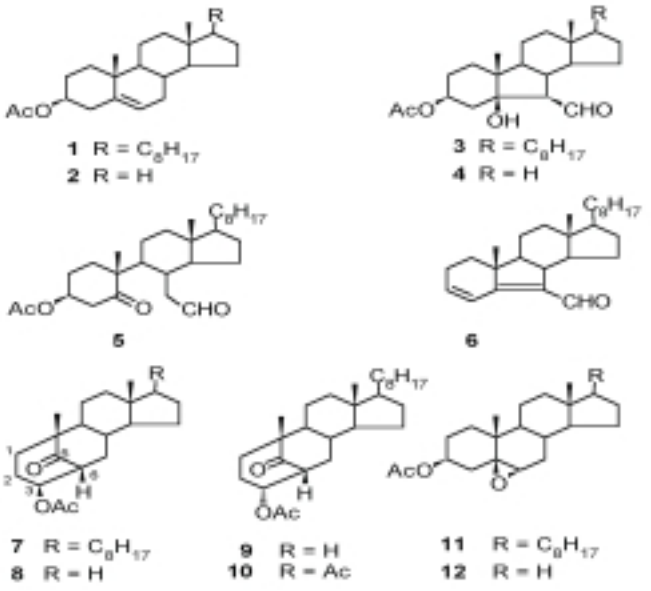
in solvents containing water or alcohols, the solvent participated products, for example 5α-hydroperoxy-7α-alkoxy-5α-B-homo-6-oxa steroids, are formed in high yield [9].

The recent study [10] shows that carbonyl oxides formed in the course of indene ozonolysis are effectively trapped by primary amines, resulting in intramolecular cyclization and final formation of the corresponding 1,2,4-dioxazolidines. The capture of the carbonyl oxide intermediate by amine is strongly dependent on the structure of the substrate, however. These results and the previously found efficient capture of the Criegee intermediates by nucleophilic solvents [7,9,11] in the course of cholesterol ozonolysis prompted us to investigate the reaction of cholesteryl acetate with ozone in the presence of primary amines. Preliminary experiments showed that products formed in the presence of amines differ from those detected on TLC plates in the course of ozonolysis without participation. Formation of products resulting from interaction of an amine with ozonides or carbonyl oxide intermediate might be envisioned.

RESULTS AND DISCUSSION

Ozonolyses of cholesteryl acetate were carried out in methylene chloride, diethyl ether or hexane solutions at –78°C in the presence of the following primary amines: *t*-butyl-, benzyl- and cyclohexylamine. For comparison, aniline and pyrrolidine were also used. The isolated yields of the two principal products **A** (**7**) and **B** (**3**) are given in Table 1.



The most polar product **B** of ozonolyses was suspected to be B-nor-hydroxy-aldehyde **3**. This compound was previously isolated in low yield (8%) in the course of ozone action on **1** in CH_2Cl_2 solution followed by zinc reduction and chromatography on alumina [12]. It has been reported, that when the crude ozonolysis product from **1** was treated with dimethyl sulfide, the quantitative yield of seco-ketoaldehyde **5** [13,14] was obtained. The structure **3** of substance **B** was confirmed by its spectral data. ^1H and ^{13}C NMR, IR and mass spectra were generally in agreement with those published [12,14,15]. Substance **B** upon action of p-toluenesulfonic acid in benzene gave unsaturated aldehyde **6** [12,13]. The identity of **B** was finally confirmed, when seco-ketoaldehyde **5** prepared in the reduction of 3β-acetoxy-5α-hydroperoxy-7α-methoxy-5α-B-homo-6-oxacholestane [9] was subjected to aldol condensation on silica gel to give compound identical with **3**.

Table 1. Products of ozonolysis of cholesteryl acetate **1** at –78°C in the presence of amines.

Amine	Solvent	Priducts[a]			Work-up conditions[c]
		3	**7**	**11**[b]	
t-$BuNH_2$	$CHCl_3$	54	18		A
	Et_2O	32	21	7	A
$PhCH_2NH_2$	CH_2Cl_2	30	26	10	B
	hexane	17	20		B
Cyclohexylamine	CH_2Cl_2	26	36	4	B
		27	42	3	C
Aniline	Et_2O	41		5	A
Pyrrolidine	Et_2O	43		6	B

[a]% yield of isolated compounds.
[b]The yield was estimated from ^1H NMR spectra.
[c]A: i) extraction, ii) chromatography; B: i) evaporation of the solvent, ii) chromatography; C: i) the reaction solution was left for 48 h at r. temp., ii) evaporation of the solvent, iii) chromatography.

Compound **A** was obtained as crystalline substance of m.p. 151–153°C, whose mass spectrum exibited molecular ion at m/z = 430 indicating the loss of one carbon atom from the substrate molecule. The IR spectrum of **A** showed bands at 1731 and 1711 cm^{-1}, which could be assigned to acetyl and 6-membered ring carbonyl groups. This was confirmed by the ^{13}C NMR spectrum, which showed two low frequency signals at δ 218 for 6-membered carbonyl group and at δ 170 for acetyl carbonyl. The ^1H NMR spectrum of **A** showed only one low frequency narrow signal ($w_{1/2}$ = 7.2 Hz) at δ 5.15, suggesting the equatorial position of proton at C(3). Consequently, inversion of the ring A chair conformation might be assumed. Additionally, the IR absorption at 1711 cm^{-1} assigned to a ketone carbonyl excluded 3β-acetoxy-B-nor-cholestan-6-one [16] as the product **A**. All these spectroscopic data were consistent with structure **7,** which is rather unusual for the product of cholesterol ozonolysis. When compound **7** was treated with potassium hydroxide in ethanol at room temp. for 24 h, hydrolysis of the acetate group was accompanied by partial isomerization at C(3). The ^1H NMR

spectrum of inseparable mixture of isomers showed a multiplet of 6β-proton at δ 2.83 and a broad signal of 3β-proton at δ 3.86 assigned to isomer **9** [17a]. Upon acetylation the latter signal moved to δ 4.85 in compound **10** [18]. The proportion of 3β-OAc and 3α-OAc isomers **7** and **10** estimated from the integration of relevant signals in ^{1}H NMR spectrum at δ 5.15 and 2.64 for **7** and δ 4.85 and 2.94 for **10** was approx. 1:1.2. Formation of the mixture of 3-epimers of **7** presumably arises by retro-aldol ring opening and subsequent reclosing reaction leading to A,B-ring bicyclic structure with the secured 6β-configuration. Formation of the 6α-isomer of **7** (as 3β-OH) or **9** is excluded for steric reasons.

Besides the two major products **3** and **7**, isolated from ozonolysis of cholesteryl acetate, a third one, the 5β,6β epoxide **11** [19] was formed in variable amounts, not exceeding 10% isolated yield, which varied slightly with reaction conditions and isolation procedure. The epoxide **11** could hardly be separated from **7**, because of similar polarity, however, it was easily detected in ozonolysis mixtures by characteristic signal in the ^{1}H NMR spectrum, which appeared as a doublet (J = 2.2 Hz) at δ 3.08 [19]. Neither the epoxide **11** nor its 5α,6α isomer was detected among products of cholesterol ozonization in nonparticipating solvents [11].

The structure **7** of the unusual anomalous ozonolysis product **A** was finally confirmed by X-ray analysis. However, the crystals were poorly diffracting and showed some signs of disorder on the side of the C(17) substituent. Therefore, although we were able to solve the structure, it was apparent that the number of observed reflections was too low to successfully refine it. It was hoped that a derivative lacking the side chain would give crystals suitable for satisfactory measurements. For this purpose, the steroid **2** lacking the side chain was subjected to ozonolysis. The following compounds were isolated: the epoxide **12** [20], the abeo-compound **8** and B-nor hydroxyaldehyde **4** [21] in 9, 26 and 23% yield, respectively. These structures were deduced on the basis of characteristic spectral data (see Experimental) which were compared with those of the analogous compounds obtained from cholesteryl acetate.

Compound **8** was subjected to X-ray analysis. X-ray data for this compound were of significantly higher quality than for **7**. Crystal data and details of the X-ray measurements are summarized in Table 2. Table 3 lists endocyclic torsion angles. The perspective view of the molecule is shown in Fig. 1. The four-ring framework contains oxo-bridged rings A and B and *trans* fused rings B/C and C/D. The stereochemistry of the four-ring skeleton is, thus, *cis-transoid-trans-transoid-trans.* A search of the Cambridge Structural Database [22] for the A-nor androstane subunit containing bridged A/B rings yielded no hits. Thus, the presented structure is the first steroidal analogue of the A-nor-3(5→6α)abeo-6β(H)-androstan-5-one type determined by X-ray diffraction. The six-membered A and C rings have chair conformations with the average torsion angle moduli 55.4(5.7) and 54.1(3.3)°, respectively. The five-membered D ring has a 13β,14α half-chair conformation.

Table 2. Crystal-data and details of X-ray measurements for **8**.

Empirical formula	$C_{20}H_{30}O_3$
Formula weight	318.44
Temperature	293 K
Wavelength	CuK_α (1.54178 Å)
Crystal system	Orthorhombic
Space group	$P2_12_12_1$
Unit cell dimensions	a = 6.748(1) Å
	b = 10.886(2)
	c = 24.185(5)
Volume	1776.6(6) Å^3
Z	4
Density (calculated)	1.191 gcm^{-1}
Absorption coefficient	0.615 mm^{-1}
Crystal size	0.32 × 0.24 × 0.12 mm
θ_{max} for data collection	65.05°
Index ranges	$-7 \le h \le 7$
	$0 \le k \le 12$
	$0 \le l \le 28$
Reflections collected	2985
Observed reflections [$F > 4\sigma(F)$]	2437
Extinction parameter	0.0069(5)
Weighting scheme	$W = 1/[\sigma^2(F_O)^2 + (0.0514P)^2 + 0.4109P]$
	$P = (maxF_o^2 + 2F_c^2)/3$
Goodness-of-fit	1.015
Final R indices (observed data)	R_1 = 0.0357
	wR_2 = 0.0940
Largest diff. peak	0.148
and hole	–0.147 e Å^{-3}

Table 3. Endocyclic torsion angles for **8** (°).

Ring A	C(10)–C(1)–C(2)–C(3)	50.3 (3)
	C(1)–C(2)–C(3)–C(6)	–48.3 (3)
	C(2)–C(3)–C(6)–C(5)	53.2 (2)
	C(3)–C(6)–C(5)–C(10)	–63.2 (2)
	C(6)–C(5)–C(10)–C(1)	62.7 (2)
	C(5)–C(10)–C(1)–C(2)	–54.6 (2)
Ring B	C(10)–C(5)–C(6)–C(7)	59.8 (2)
	C(5)–C(6)–C(7)–C(8)	–3.1 (3)
	C(6)–C(7)–C(8)–C(9)	–53.6 (2)
	C(7)–C(8)–C(9)–C(10)	57.8 (2)
	C(8)–C(9)–C(10)–C(5)	–5.0 (2)
	C(9)–C(10)–C(5)–C(6)	–54.7 (2)
Ring C	C(9)–C(8)–C(14)–C(13)	54.4 (2)
	C(8)–C(14)–C(13)–C(12)	–58.5 (3)
	C(14)–C(13)–C(12)–C(11)	57.1 (3)
	C(13)–C(12)–C(11)–C(9)	–55.0 (3)
	C(12)–C(11)–C(9)–C(8)	50.5 (3)
	C(11)–C(9)–C(8)–C(14)	–49.1 (2)
Ring D	C(13)–C(14)–C(15)–C(16)	–37.0 (2)
	C(14)–C(15)–C(16)–C(17)	12.8 (3)
	C(15)–C(16)–C(17)–C(13)	16.0 (3)
	C(16)–C(17)–C(13)–C(14)	–37.6 (3)
	C(17)–C(13)–C(14)–C(15)	46.4 (2)

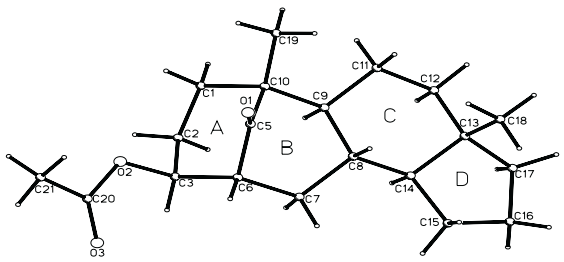


Figure 1. Perspective view of molecule **8**

The oxo-bridge constrains the cyclohexane B-ring in a boat conformation. The average torsion angle moduli within this ring amounts to 39.0(24.8)°. Ozonolysis of **1** in diethyl ether solution in the presence of aniline or pyrrolidine gave **3** in approx. 40% yield and epoxide **11** in low yield (Table 1). A series of experiments has shown, that compounds **3** and **7** were formed in various proportions in the course of ozonolysis of cholesteryl acetate carried out in Et_2O or $CHCl_3$ solutions in the presence of selected primary amines. The ^{1}H NMR spectrum of the crude ozonolysis product, taken immediately after evaporation of the solvent, is quite complex and its full analysis was impossible. This sample after standing for two days at room temperature clearly showed characteristic signals of the C(18)-methyl group found in the spectra of pure compounds **3, 5, 7** and **11** at δ: 0.73, 0.68, 0.61 and 0.64, respectively, as well as other expected low field signals. The above mentioned high frequency signals were not present in the ^{1}H NMR spectrum of the crude product, resulting from ozonolysis carried out in the absence of the amine. Instead, in this case a complex pattern of signals centered at δ 0.65 was observed. It is also clear that aldol condensation of the seco-aldehyde **5** to compound **3** occurs partially before chromatographic separation and that the process goes to completion on SiO_2 column. Thus, when pure seco-aldehyde **5** was subjected to SiO_2 column which was eluted after several hours, the eluate contained only compound **3.** Interestingly, when ozonolysis was carried out without added amine and the crude reaction product was separated on SiO_2 column, a complex mixture of several substances of polarity similar to **3** was isolated. However, the ^{1}H NMR spectrum excluded presence of **3** in significant concentration. Thus, it appears, that the added amine participates in decomposition of ozonides. When ozonolysis mixture was left for two days at room temperature in CH_2Cl_2 solution, its ^{1}H NMR spectrum shows signals indicating the presence of compound **7** and the absence of **3** and **5.** This result shows, that the primary ozonide **C** or the Criegee intermediates **D** or **E** (Scheme) reacts with the amine to give **7,** while secondary ozonide F* is stable enough [12] in solution at room temperature and decomposes partially to

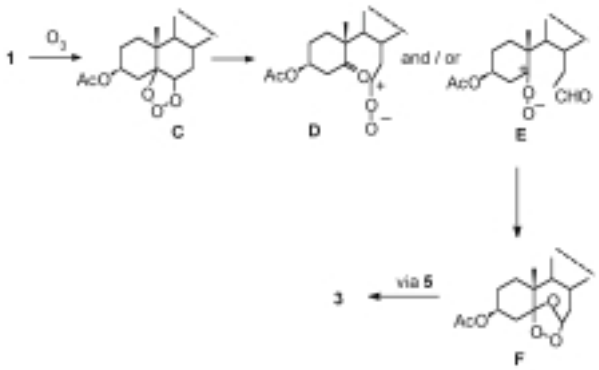
*Cholesterol ozonides (1,2,4-trioxolanes) were isolated by HPLC separation; were found unstable and decomposed to unidentified products within few days [11].

seco keto-aldehyde **5** after evaporation of the solvent (vide supra) and completely on SiO_2 column, where it undergoes aldol condensation to **3**.

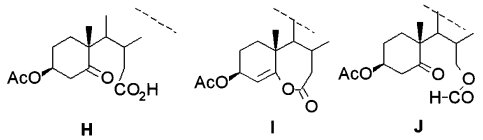
The best isolated yield of compound **7** (42%) was achieved in ozonolysis of **1** carried out in methylene chloride in the presence of cyclohexylamine, provided the crude ozonolysis mixture was left for two days at room temperature in solution allowing for decomposition of the primary ozonide **C** with amine participation. Chromatographic separation of this mixture gave **7** along with **3** (27% yield) and **11** (3%).

The reaction of cholesteryl and androstenyl acetates with ozone in amine containing solvents represents another case of „anomalous ozonolysis" resulting in rupture of the C-C bond next to the site of unsaturation. The product of anomalous ozonolysis* of cholesterol resulting from migration of C(5)–C(10) bond to peroxidic oxygen atom has been observed previously [7].

Scheme



While the mechanism of formation of 3(5→6α)abeo steroids in the course of ozonolysis is not known, the loss of C(6)-carbon atom as the formate seems conceivable.



The possible intermediate **H** or its cyclization product **I**, which would indicate abstraction of H(6) from 1,2,4-trioxolane **F**, and ketoformate **J** (migration of C(6)–C(7) bond to peroxidic oxygen) were not observed in the reaction mixtures. Furthermore, no product indicating incorporation of an amine nitrogen atom to peroxidic ozonolysis intermediates was found throughout the investigation.

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*Abnormal ozonolysis products were found in reactions of allylic compounds, α,β-unsaturated ketones, dienones and others [2b].

A-nor-3(5→6)abeo-steroids were obtained for the first time by Martin *et al*. [17a] in the $LiAl(t\text{-}BuO)_3H$ reduction of Turner`s enol lactone [23,24]. Similar transformations of androstane derivatives have also been described [17b]. Thus, the previously reported synthesis of this unusual transformed steroidal skeleton required four step transformation of Δ^5-unsaturated steroids. Compounds with A/B ring structure having bicyclo[3,3,1]nonane nucleus were found in limonoids [25].

In summary, the new route to the bridged-A-B-ring nor-steroid skeleton by ozonolysis of 3β-acetoxy-Δ^5-steroids in the presence of amines is presented. The structure of the transformed steroid is confirmed by X-ray analysis. This appears to be the first crystallographic analysis of the steroid having A-nor-3(5→ 6)abeo skeleton.

EXPERIMENTAL

M.p. values were determined on a Kofler hot-stage apparatus and are uncorrected. IR Spectra were determined with a FT-IR Bruker FS 113V spectrophotometer. ^1H and ^{13}C NMR Spectra were recorded with a Varian Gemini 300 VT spectrometers operating in the Fourier transform mode using solutions in deuteriochloroform. Coupling constants J are given in Hz, and the chemical shifts (δ) are expressed in ppm relative to tetramethylsilane. Electron impact mass spectra were recorded with a AMD 402 spectrometer. Column chromatography was performed by using silica gel 60 (Merck 70–230 mesh, no. 7734). The progress of reactions was monitored by TLC using a precoated aluminium-backed silica plates (E. Merck, no. 5554).

X-ray analysis and structure refinement: Single crystals of **8** were obtained from a solution of the compound in methanol. The reflection intensities were measured on a four-circle KM-4 (KUMA DIFFRACTION) diffractometer equipped with graphite monochromator. The reflections were measured using ω-2θ scan technique with variable scan rate, and a scan range in ω 0.9°. Background measurements were estimated from 68-step profile. The intensities were corrected for Lorentz and polarization effects, absorption corrections were not applied. The structures were solved by direct methods with SHELXS-86 [26] and refined with SHELXL-97 [27]. Heavy atoms (C, O, N) were refined anisotropically. The positions of the H-atoms were calculated and refined using a riding model with an isotropic temperature factor 1.3 times U_{eq} of the atom to which they are bonded. The absolute structure of the crystals was assumed from the known absolute configuration of cholesteryl acetate. Siemens computer graphics program [28] was used to prepare drawings. Details of the present X-ray analysis and crystal data are collected in Table 2 and 3*.

The general procedure of ozonolysis of 1 in presence of an amine: Cholesteryl acetate **1** (300 mg) was dissolved in a solvent (15 ml), amine (5 eq.) was added and the solution cooled to –78°C. The oxygen was passed through the solution followed by stream of ozone until all the substrate disappeared (TLC test, KI test for excessive ozone). The solution was flushed with oxygen and was warmed slowly to room temperature. Evaporation of the solvent gave the crude reaction product as an oily residue. This was usually left for several hours and was separated on SiO_2 column eluted with benzene-ethyl acetate mixtures. The compounds were eluted in the order: **11, 7** and **3**. The results are summarized in Table 1.

3: m.p. 90–92°C (from MeOH) (lit. [12] m.p. 93.5–95°C); ν_{max}/cm^{-1} (KBr) 3474; 2952; 2934; 2820; 2718; 1723; δ_H: 9.68 (1H, d, J = 3.02 Hz, CHO); 5.12 (1H, m, 3α-H); 2.49; 2.18; 2.07 (3H, s, AcO); 0.95 (3H, s, 10-Me); 0.728 (3H, s, 13-Me); *m/z*: (FAB) 463 (MH^+); 399; 383.

7: m.p. 151–153°C (from $CHCl_3$-MeOH). (lit. [18] m.p. 147–148°C); ν_{max}/cm^{-1} (KBr) 2951; 2867; 1731 (CO); 1711 (CO); δ_H: 5.15 (1H, br s, $w_{1/2}$ = 7.2 Hz, 3α-H); 2.64 (1H, dt, J_1 = 11.7, J_2 = 6.8 Hz, 6β-H);

*Atomic coordinates, anisotropic displacement parameters and tables of bond distances and angles have been deposited at the Cambridge Crystallographic Data Centre (Dep. No. CCDC 170497).

2.01 (3H, s, OAc); 1.00 (3H, s, 10-Me); 0.61 (3H, s, 13-Me); δ_C: 218.1; 170.2; 78.5; 71.3; *m/z*: 430; 370; 342; (HR) 430.34421. Calcd. for $C_{28}H_{46}O_3$ 430.34470.

11: m.p. 110–112°C (lit. [19] m.p. 110°C); δ_H: 4.77 (1H, m, 3α-H); 3.08 (1H, d, J = 2.2 Hz, 6β-H); 2.03 (3H, s, AcO); 1.00 (3H, s, 10-Me), 0.654 (3H, s, 13-Me) (the spectrum was identical with that of an authentic sample).

Ozonolysis of olefin 2: Olefin **2** (315 mg, 0.99 mmol) was ozonized at –78°C in CH_2Cl_2 (21 ml) solution with cyclohexyl amine (0.54 ml, 4.77 mmol) for 45 min. Evaporation of the solvent and chromatograhpy on SiO_2 column afforded the following pure compounds: epoxide **12**: (29 mg, 9% yield); m.p. 97–99°C (from heptane), lit. [12] m.p. 98–100; δ_H: 4.76 (1H, m, 3α-H); 3.08 (1H, d, 6β-H); 2.01 (3H, s, AcO); 1.01 (3H, s, 10-Me); 0.66 (3H, s, 13-Me); (the ^{1}H NMR spectrum was identical with that of the original sample);

compound **8**: (82 mg, 26% yield); m.p. 165–167°C (from methanol); ν_{max}/cm^{-1} (KBr) 2952; 2874; 1732 (CO); 1717 (CO); δ_H: 5.16 (1H, m, 3α-H); 2.65 (1H, dt, 6β-H); 2.42 (1H, m); 2.16 (1H, dt); 2.03; 2.01 (3H, s, AcO); 1.01 (3H, s, 10-Me); 0.65 (3H, s, 13-Me); δ_C: 218.0 (C=O); 170.1 (AcO); 78.1 (C-3); *m/z*: 318; 258; 230; 135; 95; *m/z* (HR) 318.22203, calcd. for $C_{20}H_{30}O_3$ 318.21933;

compound **4** [21]: (oil, 73 mg, 23% yield); ν_{max}/cm^{-1} (KBr) 2718 (CHO); 1723 (CO); ν_{max}/cm^{-1} ($CHCl_3$) 2952; 2868; 1717 (CO); δ_H: 9.69 (1H, d, CHO); 5.13 (1H, m, 3α-H); 2.52 (1H, s, 5-OH); 2.20; 2.07 (3H, s, AcO); 0.96 (3H, s, 10-Me); 0.77 (3H, s, 13-Me); δ_C: 203.8 (CO); 169.7 (AcO); 83.6; 64.5; 70.3; *m/z*: 348 (M^+); 288; 242; 137; *m/z* (HR) 288.20769, cacld. for $C_{19}H_{28}O_2$ 288.20892.

Isomerization of compound 7: Compound **7** (40 mg) was dissolved in 1N solution of potassium hydroxide in ethanol (7 ml) and the mixture was left at room temperature for 24 h. H_2O was added and the product was extracted with benzene-ether (1:1) mixture. The crude product (39 mg), which was a mixture of isomer **7** [as 3β-OH: δ_H: 4.21 (1H, br s, $w_{1/2}$ = 7.2 Hz, 3α-H); 2.61 (1H, dt, J_1 = 11.5, J_2 = 6.3 Hz, 6β-H); 0.90 (3H, s, 10-Me); 0.61 (3H, s, 13-Me) and isomer **9** [(δ_H: 3.80–3.92 (1H, m, 3β-H); 2.86–2.81 (1H, m, 6β-H); 0.94 (3H, s, 10-Me); 0.62 (3H, s, 13-Me)], was dissolved in pyridine (1 ml) and acetic anhydride (1 ml) and the solution was left at room temperature for 1 h. The usual work up gave the product (31 mg) as a mixture of the acetate **7** [δ_H: 5.15 (1H, br s, $w_{1/2}$ = 7.2 Hz, 3α-H); 2.64 (1H, dt, J_1 = 11.7 Hz J_2 = 6.8 Hz); 2.01 (3H, s, OAc); 1.00, 0.61] and the epimer **10** [δ_H: 4.85 (1H, dt, J_1 = 4.4, J_2 = 11.2 Hz, 3β-H); 2.94 (1H, dt, J_1 = 4.4 Hz J_2 = 10.7 Hz, 6β-H); 2.05 (3H, s, OAc); 0.95 (3H, s, 10-Me); 0.62 (3H, s, 13-Me)] in the ratio 1:1.2.

The reaction of seco-ketoaldehyde 5 on silica gel column: A solution of compound **5** (40 mg) in benzene (1.5 ml) was introduced to SiO_2 (2 g) column and left at room temperature for 36 h. Elution with benzene-ethyl acetate (60:1) gave **3** (29 mg), whose ^{1}H NMR spectrum was identical with that of the original sample.

Action of *p*-toluenesulfonic acid on compound 3: To a solution of compound **3** (51 mg) in benzene (2.5 ml) *p*-TsOH (3.2 mg) was added and the mixture was stirred at 50°C for 6 h. After cooling, the benzene solution was washed with aq. $NaHCO_3$ (5%), water and brine, dried ($MgSO_4$) and evaporated. The crude residue (36 mg) dissolved in benzene was filtred through SiO_2 column. The pure compound **6** (oil) [12,13] had: δ_H: 10.03 (1H, s, CHO); 6.92 (1H, br d, J = 10.4 Hz); 6.22 (H, br d); 0.95 (3H, 10-Me); 0.75 (3H, s, 13-Me); δ_C: 189.7 (CHO); 163.8 (C-5); 138.3 (C-3); 136 (C-6); 120.8 (C-4); UV λ_{max} 291.6 (ε 15240); *m/z*: (FAB) 383 (MH^+).

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REFERENCES

1. Bailey P.S., in *Ozonation in Organic Chemistry*, Academic Press, NY, vol. 2, Ch. 7 (1982).
2. Bailey P.S., in *Ozonation in Organic Chemistry*, Academic Press, NY, vol. **1**, a) Ch. 7, b) Ch. 9 (1978).
3. Fremery M.I. and Fields E.K., *J. Org. Chem.*, **29**, 2240 (1964).
4. Hon Y.S., Lin S.W., Lu L. and Chen Y.J., *Tetrahedron*, **51**, 5019 (1995).

5. Schulz M., Becker D. and Rieche A., *Angew. Chem*. *Int. Ed*., 548 (1965).
6. Aurell M.J., Ceita L., Mestres R. and Tortajada A., *Tetrahedron*, **53**, 10883 (1997).
7. Gumulka J. and Smith L.L., *J. Am. Chem. Soc*., **105**, 1972 (1983); Smith L.L., Ezell E.L. and Jaworski K., *Steroids*, **61**, 401 (1996).
8. Smith L.L., *Lipids*, **31**, 453 (1996) and references cited therein.
9. a) Paryzek Z., Martynow J. and Swoboda W.*, J. Chem. Soc., Perkin Trans. 1*, 1222 (1990); b) Paryzek Z. and Rychlewska U., *J. Chem. Soc., Perkin Trans. 2*, 2313 (1997) and references cited therein.
10. Ushigoe Y., Satake S., Masuyama A., Nojima M. and McCullough K.J., *J. Chem. Soc., Perkin Trans. 1*, 1939 (1997).
11. Jaworski K. and Smith L.L., *J. Org. Chem*., **53**, 555 (1988).
12. Tanabe K., Hayashi R. and Takasaki R., *Chem. Pharm. Bull*., **9**, 1 (1961).
13. Yates P. and Stiver S., *Can. J. Chem*., **66**, 1209 (1988).
14. Morand P. and Kaufman M., *J. Org. Chem*., **34**, 2175 (1969).
15. Dave V. and Stothers J.B., *Can. J. Chem*., **57**, 1550 (1979).
16. Morisawa Y., *Chem. Pharm. Bull*., **12**, 1066 (1964).
17. a) Martin J., Parker W., Shroot B. and Steward T., *J. Chem. Soc. C*, 101 (1967); b) Kasal A., *Collect. Czech. Chem. Commun*., **43**, 1778 (1978).
18. Snatzke G. and Kinsky K., *Tetrahedron*, **28**, 289 (1972).
19. Marchon J-C. and Ramasseul R., *Synthesis*, 389 (1989).
20. Hanson J.R. and Terry N., *J. Chem. Res. (S)*, 50 (1998).
21. Nagata W., Narisada M., Wakabayashi T., Hayase Y. and Masayuki M., *Chem. Pharm. Bull*., **19**, 1567 (1971).
22. Allen F.H. and Kennard O., *Chemical Design Automation News,* **8***,* 1**&**31–37 (1993).
23. Turner R.B., *J. Am. Chem. Soc*., **72**, 579 (1950).
24. Weill-Raynal J., *Synthesis*, 49 (1969) and references cited therein; Julia S.A., Eschenmoser A., Heuser H. and Tarkey N., *Helv. Chim. Acta*, **36**, 1885 (1953).
25. Kehrli A.R.H., Taylor D.A.H. and Niven M., *J. Chem. Soc., Perkin Trans. 1*, 2057 (1990).
26. Sheldrick G.M., *Acta Cryst.,* **A46**, 467 (1990).
27. Sheldrick G.M., SHELXL-97, University of Göttingen, (1997).
28. *Stereochemical Workstation Operation Manual*, Release 3.4, Siemens Analytical X-Ray Instruments, Inc., Medison, Wisconsin, USA, (1989).