SYNTHESIS, REACTIONS, CONFORMATION ANALYSIS, AND NMR SPECTRA OF 5,10-EPOXY-5ξ,10ξ-ESTRANE-3,17-DIONES+

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On epoxidation of estr-5(10)-ene-3,17-dione, the 5β,10β-epoxide is the major product. The mixture of epoxides was converted into 5,10β-dihydroxy-5α-estrane-3,17-dione, 10α- and 10β-hydroxyestr-4-ene-3,17-dione, and estrone. Due to the sensitivity of the products, ^{1}H NMR was the best way to monitor the reaction pathway.

Keywords: Steroids; Epoxidation; Epoxide cleavage; Stereoselective reactions; Conformation analysis; NMR spectroscopy.

The removal of the angular methyl group at C-10 of certain steroids has led to a marked increase in biological activity. This is particularly apparent in 17α-ethynyl-19-nortestosterone whose high oral hormonal activity has led to the introduction of that compound into medical practice (norlutin). Recently, renewed interest was paid to estr-5(10)-ene-3,17-dione (**1**; Scheme 1) the analytical standards of which, as well as those of some related derivatives, were needed. This is why we decided to study the reactivity of the compound **1**.

As early as 1954, Colton[2] patented the epoxidation of olefin **1**; however, he isolated no epoxides (i.e., compounds **2** and **3**) but products of their isomerisation only: on treatment with sodium carbonate, the primary products – epoxides **2** and **3** – yielded two unsaturated alcohols supposed to be 10ξ-hydroxyestr-4-en-3-ones (i.e., compounds **4** and **5**, respectively). The similarity of the physical data of one of the epoxides (**2**) to those of 10β-hydroxyestr-4-ene-3,17-dione[3] (**4**), obtained by microbial hydroxylation of 19-nortestosterone, suggested the 10β-hydroxy structure for the

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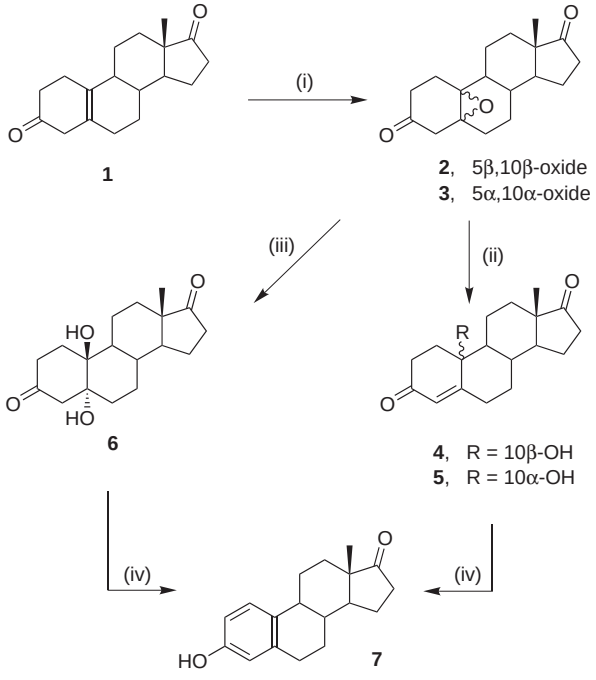
major alcohol **4**. This points to the 5β,10β-epoxide **2** as the major component of the epoxide mixture. Eventually, Djerassi[4] proved the 10β-configuration of the major 10-alcohol **4** using optical rotatory diffraction: the ORD curve was close to that of testosterone, the 10α-alcohol **5** would give an antipodal effect.



(i) peroxyphthalic acid; (ii) AcOH, heating; (iii) H^+ or silica gel; (iv) silica gel

SCHEME 1

The 5α,10α-epoxide **3** has never been isolated. Chemical Abstracts contain no reference to it although related derivatives (i.e., compounds with a sp^3 hybridisation at C-17 carbon) have been described[5,6]. Traditional chromatographic separation of the epoxides led to the above products of the oxirane ring opening, compounds **4** and **5**. Adsorbents used were plain or ammonia treated silica gel, aluminium oxide, and the reverse silica based phase (C-18). Eluent used was either chloroform (as obtained and washed with ammonia) or aqueous methanol. Thin layer chromatography, column chromatography, high-performance liquid chromatography (HPLC) have been used to no avail. Analytical HPLC was rapid enough not to destroy the epoxides; however, preparative HPLC again yielded the products of the

ring-opening. Neither the 10α-alcohol **5**, apart from Colton[2], has ever been isolated. We also succeeded in its isolation and found its properties identical with those described by Colton for the minor product of epoxidation.

The predominance of the 5β,10β- over 5α,10α-epoxide in epoxidation of the $\Delta^{5(10)}$-double bond is surprising in view of the generally known better accessibility for the α-approach of reagents to the steroid molecule. Djerassi's[4] explanation of the stereochemistry of epoxidation is that the 5β,10β-epoxide **2** is close to the normal (5β,9α,10β) configuration of most steroid compounds while 5α,10α-epoxide involves the unfavourable 9,10-syn configuration of the A and B rings.

The epoxide cleavage was optimised for preparative purposes.

Various acids were used and sulfuric acid in dioxane was found the best to produce 5,10β-dihydroxy-5α-estrane-3,17-dione (**6**) in addition to compound **4**. When the latter compound was sought, the best procedure was found to be heating the solution of epoxides in anhydrous acetic acid. Since the prolonged heating of the mixture produced estrone (**7**), care had to be taken in the work-up of the mixture. To avoid the estrone formation, acetic acid had to be removed carefully by dilution of the reaction mixture with chloroform and washing the solution with water and aqueous sodium carbonate.

The 5α,10β-diol **6** was also dehydrated to the unsaturated dione **4** either by heating or action of acidic conditions. The ease of dehydration was also the reason for the large discrepancy among reports on its melting point: at a fast rise of temperature, the m.p. was found at 222–228 °C, at a slow rise, the value became higher (ca. 236–241 °C), i.e. closer to the melting point of estrone (254–255 °C); TLC showed the prevalence of estrone (**7**) in the sample melted.

In summary, all transformations of the epoxides **2** and **3** were consistent with previous knowledge of epoxide hydrolysis: in agreement with the Fürst–Plattner[7] rule, the 5β,10β-epoxide **2** yielded the 10β-alcohols **4** and **6** where the hydroxy group is axial within the B ring while the 5α,10α-epoxide **3** produced the diaxial diol **5** and the unsaturated 10α-alcohol **6** where the 10α-hydroxy group is axial within the A ring.

NMR DISCUSSION

The structures of compounds **1**–**7** were confirmed by detailed analysis of ^1H and ^{13}C NMR spectra in $CDCl_3$ and C_6D_6. When $CDCl_3$ is used as a solvent then the traces of acid have to be removed (e.g. with alkaline Al_2O_3) before NMR measurement. For monitoring the quality of the above products C_6D_6

is probably the safer solvent. Proton and carbon chemical shifts in both $CDCl_3$ and C_6D_6 are given in Tables I and II.

TABLE I
^{1}H NMR chemical shifts of compounds **1**–**7** in $CDCl_3$ and C_6D_6 (in parentheses)

Proton	**1**	**2**	**3**	**4**	**5**	**6**[a]	**7**
1α	2.08	2.45	1.98	1.96	2.11	1.98	7.15
	(2.08)	(1.98)	(1.83)	(1.40)	(1.34)	(2.21)	(7.10)
1β	1.98	1.98	1.47	2.21	1.95	2.10	–
	(1.91)	(1.40)	(1.48)	(1.59)	(1.56)	(1.92)	
2α	~2.47	2.37	2.22	2.34	2.75	2.61	6.64
	(2.35)	(2.34)	(2.46)	(2.21)	(2.59)	(2.39)	(6.59)
2β	~2.47	2.27	1.98	2.57	2.47	2.31	–
	(2.20)	(2.10	(2.09	(2.25)	(2.38)	(2.71)	
4α	2.80	2.83	2.78	5.77	5.83	3.03	6.59
	(2.63)	(2.70)	(2.63)	(5.84)	(5.86)	(2.31)	(6.49)
4β	2.69	2.61	2.57	–	–	2.09	–
	(2.57)	(2.21)	(2.24)			(3.14)	
6α	2.47	1.83	1.67	2.35	2.63	1.41	~2.87
	(1.66)	(1.36)	(1.14)	(1.94)	(1.72)	(1.52)	(~2.74)
6β	2.43	2.00	2.11	2.69	2.21	2.01	~2.87
	(1.75)	(1.75)	(1.78)	(2.33)	(2.21)	(2.12)	(~2.74)
7α	1.85	1.62	2.41	1.14	1.60	1.40	1.44
	(1.02)	(0.62)	(1.01)	(0.68)	(0.97)	(1.49–1.55)	(1.18)
7β	1.31	1.03	2.07	1.98	1.89	1.63	2.00
	(1.56)	(1.26)	(1.50)	(1.53)	(1.31)	(1.49–1.55)	(1.71)
8	1.49	1.63	1.22	1.97	1.35	1.74	1.57
	(1.20)	(1.50)	(0.79)	(1.65)	(0.73)	(1.82)	(1.32)
9	1.75	1.38	1.57	1.14	1.75	1.57	2.24
	(1.39)	(0.92)	(1.33)	(0.58)	(1.25)	(1.67)	(2.03)
11α	1.98	1.94	1.36	1.78	1.83	1.72	2.38
	(1.60)	(1.58)	(1.31)	(1.29)	(1.52)	(1.54–1.60)	(2.16)
11β	1.25	1.56	1.21	1.70	1.57	1.42	1.50
	(0.93)	(1.33)	(1.06)	(1.38)	(1.13)	(1.54–1.60)	(1.35)
12α	1.36	1.36	1.55	1.31	1.30	1.36	1.47
	(1.33)	(1.31)	(1.29)	(1.21)	(1.20)	(1.34)	(1.44)
12β	1.86	1.92	1.90	1.89	1.89	1.89	1.96
	(1.95)	(1.97)	(1.91)	(1.94)	(1.93)	(1.96)	(2.04)
14	1.45	1.36	1.33	1.32	1.34	1.42	1.52
	(1.06)	(0.95)	(0.87)	(0.85)	(0.76)	(1.17)	(1.05)
15α	1.98	1.92	1.92	1.98	1.97	1.98	2.05
	(1.56)	(1.49)	(1.45)	(1.50)	(1.46)	(1.68)	(1.55)
15β	1.59	1.56	1.56	1.59	1.52	1.55	1.62
	(1.17)	(1.10)	(1.08)	(1.12)	(1.05)	(1.27)	(1.15)
16α	2.11	2.08	2.08	2.10	2.09	2.11	2.15
	(1.90)	(1.88)	(1.85)	(1.88)	(1.85)	(1.89)	(1.88)
16β	2.49	2.46	2.46	2.48	2.46	2.47	2.51
	(2.25)	(2.20)	(2.21)	(2.22)	(2.23)	(2.24)	(2.22)
18-Me	0.908	0.911	0.900	0.945	0.829	0.913	0.912
	(0.744)	(0.721)	(0.707)	(0.756)	(0.547)	(0.855)	(0.692)

[a] Values in parentheses for compound **6** were obtained in the mixture of C_6D_6 and CD_3COCD_3 5:1.

TABLE II
^{13}C NMR chemical shifts of compounds **1**–**7** in $CDCl_3$ and C_6D_6 (in parentheses)

Carbon	**1**	**2**	**3**	**4**	**5**	**6**[a]	**7**
1	27.33 (27.49)	24.04 (24.12)	23.56 (23.69)	33.59 (33.75)	30.48 (30.43)	32.12 (31.98)	126.48 (126.73)
2	38.93 (38.92)	35.63 (35.46)	35.36 (35.33)	33.53 (33.59)	33.11 (33.41)	37.00 (37.40)	112.83 (113.09)
3	210.94 (207.93)	207.82 (205.76)	207.73 (205.74)	199.14 (196.84)	198.98 (197.20)	210.56 (209.01)	153.52 (154.36)
4	44.52 (44.62)	45.44 (45.45)	44.14 (44.29)	124.64 (125.00)	126.64 (126.30)	51.52 (51.65)	115.28 (115.50)
5	128.60 (126.62)	60.41 (60.11)	60.80 (60.30)	164.30 (162.39)	165.25 (163.91)	73.20 (73.05)	132.10 (132.01)
6	30.47 (30.53)	28.55 (28.61)	30.35 (30.37)	31.72 (31.59)	26.76 (26.58)	34.35 (34.26)	29.43 (29.71)
7	25.63 (25.86)	24.82 (25.00)	24.08 (23.97)	30.47 (30.56)	27.99 (27.89)	24.39 (24.87)	26.47 (26.70)
8	38.53 (38.58)	33.62 (33.65)	37.93 (38.04)	34.68 (34.61)	30.86 (30.48)	34.66 (34.79)	38.37 (38.48)
9	46.07 (46.18)	46.03 (46.09)	46.31 (46.49)	52.58 (52.34)	48.61 (48.28)	45.44 (45.85)	43.96 (44.11)
10	130.62 (130.56)	63.19 (62.87)	62.67 (62.22)	70.06 (69.87)	70.89 (70.36)	77.14 (76.89)	138.01 (137.81)
11	24.63 (24.70)	22.33 (22.51)	22.16 (22.41)	19.60 (19.62)	20.34 (20.34)	20.15 (20.35)	25.92 (26.15)
12	31.73 (32.32)	31.44 (32.04)	31.72 (32.77)	30.95 (31.53)	30.74 (31.22)	31.18 (31.98)	31.58 (32.09)
13	48.16 (47.93)	47.82 (47.68)	47.75 (47.55)	47.54 (47.30)	47.66 (47.26)	47.59 (47.52)	48.00 (47.69)
14	49.94 (49.90)	49.70 (49.70)	50.17 (49.99)	50.38 (50.26)	52.87 (52.59)	50.33 (50.52)	50.45 (50.14)
15	21.34 (21.36)	21.60 (21.65)	21.35 (21.37)	21.73 (21.72)	21.72 (21.64)	21.76 (21.90)	21.56 (21.47)
16	35.76 (35.55)	35.60 (35.54)	35.60 (35.59)	35.70 (35.52)	35.50 (35.28)	35.78 (35.67)	35.84 (35.56)
17	220.61 (217.54)	220.23 (217.65)	219.88 (217.37)	220.61 (217.51)	219.86 (216.99)	220.56 (218.51)	220.91 (217.91)
18	13.97 (13.90)	13.85 (13.72)	13.90 (13.83)	13.64 (13.53)	13.25 (13.23)	13.73 (13.67)	13.83 (13.65)

[a] Values in parentheses for compound **6** were obtained in the mixture of C_6D_6 and CD_3COCD_3 5:1.

The assignment of the configuration of epoxides **2** and **3** in the mixture is not an easy task. The inspection of models shows that there are no favourable NOE contacts for distinguishing between **2** and **3**. Our assignment is based on comparison of the chemical shifts of axial protons H-7α, H-8, H-9, and H-11β, which should be influenced mainly by orientation of the epoxy ring. The observation of downfield shifts of H-8 and H-11β (0.41 and 0.35 ppm) in the major isomer should be the result of van der Waals effect[8] of the β-faced epoxy ring while downfield shift of H-7α and H-9 (0.79 and 0.19 ppm) in the minor isomer should reflect the same effect of the α-faced epoxy ring (Fig. 1).

The configuration of the tertiary hydroxy group at C(10) in isomeric 10ξ-alcohols **4** and **5** was derived from 2D-H,H-ROESY spectra in DMSO-d_6, where the signal of 10-OH group could be easily identified. The observation of NOE cross-peaks of OH (δ 5.07) with H-1β (δ 2.06), H-2β (δ 2.51), and namely H-8 (δ 1.89) indicates the 10β-OH configuration in compound **4** (Fig. 2a). A similar procedure with 10α-alcohol **5** failed – no NOE contacts of 10-OH group were observed, obviously due to faster exchange of OH proton even in DMSO-d_6. Therefore we used an *in situ* reaction (Scheme 2) of compound **5** with trichloroacetyl isocyanate (TAI) in $CDCl_3$ (for details of the TAI method, see lit.[9,10]). Significant induced TAI acylation shifts ($|\Delta\delta| >$ 0.15 ppm) were observed only for hydrogens H-2α (+0.45 ppm), H-6α (+0.73 ppm), and H-9α (+0.76 ppm) that prove 10α-configuration of the OH group in compound **5** (Fig. 2b). In the case of 5,10-diol **6** we again successfully used the 2D-H,H-ROESY spectrum (in a mixture of C_6D_6 and CD_3COCD_3 5:1) to determine the configurations at C(5) and C(10). The NOE contacts of OH proton at δ 3.01 with H-1β (δ 1.92), H-2β (δ 2.71), and H-11β (δ 1.57) suggest the 10β-OH position while NOE contacts of OH at δ 3.26 with H-4α (δ 2.31) and H-6, H-7α (both at δ ≈1.52) prove 5α-OH con-

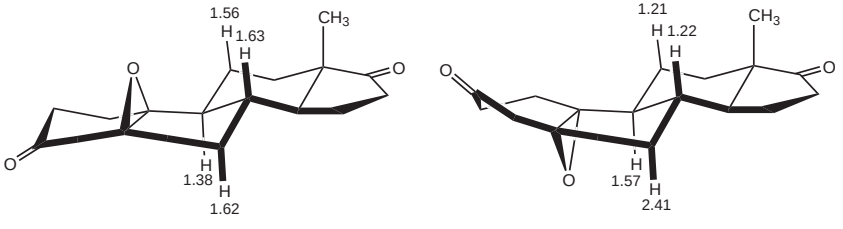


FIG. 1
Geometry of isomeric epoxides **2**, **3** and chemical shifts of H-7α, H-8, H-9, and H-11β

figuration in the diol **6** (Fig. 2c). The position 3 for phenolic OH in compound **7** follows from the coupling pattern of aromatic protons at ring A (Fig. 2d).

The easy derivatisation of tertiary alcohols with TAI, described above for the determination of configuration of the 10-OH group in ketodiol **5**, led us to the application of this procedure also to the remaining hydroxy derivatives **4**, **6**, and **7**. While the 10β-alcohol **4** provided the expected 10-OTAC derivative **4a** and estrone (**7**) gave corresponding 3-OTAC derivative **7a**, the reaction of 5,10-diol **6** with TAI was much more complex (Scheme 2). After the first addition of a small amount of TAI, two mono-TAC derivatives **6a** and **6b** in the ratio ca. 35:65 appeared (5α-OH is probably more easily accessible to TAI attack) in addition to the starting diol **6**. Further addition of TAI led first to the formation of derivative **6c** and then – after ca. 24 h standing with excess of TAI, the products of elimination – compounds **4a**

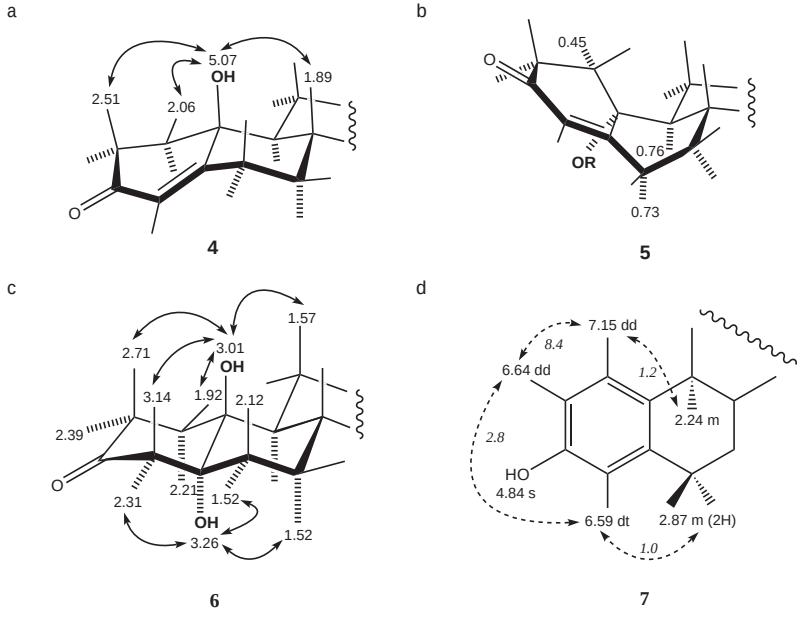
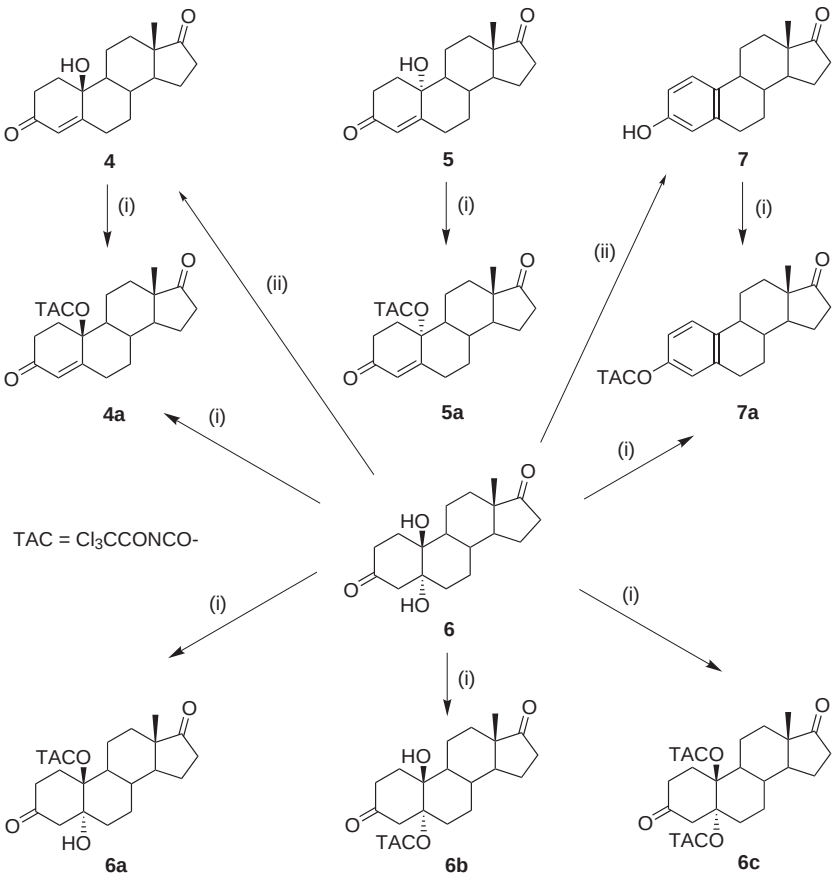


FIG. 2

The NMR arguments for the configuration of OH groups in compounds **4–7**: a the observed NOE contacts of 10-OH in **4** (indicated with arrows); b TAI acylation shifts (Δδ) of hydrogens H-1α, H-6α, and H-9α in **5**; c the observed NOE contacts of 5-OH and 10-OH in **6** (indicated with arrows); d chemical shifts, multiplicities, and coupling constants of hydrogens in ring A in **7**



SCHEME 2

In situ reactions of hydroxy derivatives **4–7** observed after addition of: (i) TAI and/or (ii) CH_3COOH to their $CDCl_3$ solutions in NMR tube

and **7a** (identical with those prepared previously by TAI-acylation of **4** and **7**) were formed. The 3-OTAC derivative **7a** with aromatic ring A was the final product present in the reaction mixture. The reaction course was monitored by repeating measurements of NMR spectra. The complete ^1H and ^{13}C NMR data of TAC derivatives **4a**, **5a**, **6c**, and **7a** together with TAI-induced acylation shifts are given in Table III.

Another reaction that was successfully monitored by NMR spectra was the transformation of diol **6** with acetic acid in a $CDCl_3$ solution: it led first to unsaturated 10β-alcohol **4** and finally to phenol **7**.

TABLE III
^{1}H and ^{13}C chemical shifts of TAC-derivatives **4a**, **5a**, **6c**, and **7a** in $CDCl_3$ and TAI-induced acylation shifts (in parentheses)

Proton	**4a**	**5a**	**6c**	**7a**	Carbon	**4a**	**5a**	**6c**	**7a**
1α	2.38 (+0.42)	2.56 (+0.45)	2.44 (+0.46)	7.32 (+0.17)	1	32.41 (–1.18)	29.33 (–1.15)	26.84 (–5.28)	126.63 (+0.15)
1β	2.45 (+0.24)	2.04 (+0.09)	3.56 (+1.46)	–	2	34.74 (+1.21)	32.62 (–0.49)	37.15 (+0.15)	118.14 (+5.31)
2α	2.36 (+0.02)	2.83 (+0.08)	2.47 (–0.14)	6.98 (+0.34)	3	197.24 (–1.90)	198.20 (+1.00)	206.42 (–4.14)	157.55 (+4.03)
2β	2.73 (+0.16)	2.51 (+0.04)	2.40 (+0.09)	–	4	126.03 (+1.39)	128.88 (+2.24)	45.02 (–6.50)	121.05 (+5.77)
4α	5.97 (+0.20)	5.86 (+0.03)	3.36 (+0.33)	6.95 (+0.36)	5	161.45 (–2.85)	161.54 (–3.71)	89.18 (+15.98)	138.44 (+6.34)
4β	–	–	2.97 (+0.88)	–	6	28.80 (–2.92)	29.12 (+2.36)	27.55 (–6.80)	29.39 (–0.04)
6α	2.33 (–0.02)	3.36 (+0.73)	2.42 (+1.01)	~2.93 (+0.06)	7	32.49 (+2.02)	26.11 (–1.88)	24.41 (+0.02)	26.26 (–0.21)
6β	2.73 (+0.04)	2.36 (+0.15)	2.70 (+0.69)	~2.93 (+0.06)	8	35.07 (+0.39)	31.45 (+0.59)	34.51 (–0.15)	37.97 (–0.40)
7α	1.22 (+0.08)	1.69 (+0.09)	1.12 (–0.28)	1.47 (+0.03)	9	53.26 (+0.68)	42.73 (–5.88)	46.62 (+1.18)	44.16 (+0.20)
7β	2.11 (+0.13)	1.82 (–0.07)	1.78 (–0.15)	2.04 (+0.04)	10	84.78 (+14.72)	84.44 (+13.55)	90.62 (+13.48)	147.62 (+9.61)
8	2.20 (+0.23)	1.48 (+0.13)	1.82 (+0.08)	1.62 (+0.05)	11	20.97 (+1.37)	20.53 (+0.19)	21.61 (+1.46)	25.76 (–0.16)
9	1.46 (+0.32)	2.51 (+0.76)	1.93 (+0.36)	2.30 (+0.06)	12	31.08 (+0.13)	30.50 (–0.24)	31.29 (+0.11)	31.58 (0.00)
11α	1.98 (+0.20)	1.84 (+0.01)	2.06 (+0.34)	2.42 (+0.04)	13	47.90 (+0.36)	47.71 (+0.05)	47.37 (–0.22)	47.90 (–0.10)
11β	1.85 (+0.15)	1.65 (+0.08)	1.36 (–0.06)	1.56 (+0.06)	14	50.56 (+0.18)	52.45 (–0.42)	50.42 (+0.09)	50.49 (+0.04)
12α	1.32 (+0.1)	1.33 (+0.03)	1.37 (+0.01)	1.50 (+0.03)	15	21.70 (–0.03)	21.72 (0.00)	21.79 (+0.03)	21.58 (+0.02)
12β	1.90 (+0.01)	1.89 (0.00)	1.89 (0.00)	1.98 (+0.02)	16	35.65 (–0.05)	35.47 (–0.03)	35.63 (–0.15)	35.80 (–0.04)
14	1.35 (+0.03)	1.43 (+0.09)	1.35 (–0.07)	1.53 (+0.01)	17	219.95 (–0.66)	219.32 (–0.54)	219.72 (–0.84)	220.39 (–0.52)
15α	1.98 (0.00)	2.03 (+0.06)	1.98 (0.00)	2.06 (+0.01)	18	13.63 (–0.01)	13.46 (+0.21)	13.64 (–0.09)	13.82 (–0.01)
15β	1.60 (+0.01)	1.52 (0.00)	1.60 (+0.05)	1.63 (+0.01)					
16α	2.09 (–0.01)	2.12 (+0.03)	2.09 (–0.02)	2.16 (+0.01)					
16β	2.46 (–0.02)	2.48 (+0.02)	2.46 (–0.01)	2.52 (+0.01)					
18-Me	1.060 (+0.115)	0.860 (+0.031)	0.860 (–0.053)	0.920 (0.008)					
NH	8.44	8.25	8.39 8.28	8.70					

EXPERIMENTAL

Melting points were determined on a melting point micro apparatus Boetius (Germany) and are uncorrected. Analytical samples were dried over phosphorus pentoxide at 50 °C/100 Pa. Optical rotations were measured in chloroform using an Autopol IV (Rudolf Research Analytical, Flanders, U.S.A.); $[\alpha]_D$ values are given in 10^{-1} deg cm^2 g^{-1}. IR spectra of chloroform solutions were recorded on a Bruker IFS 88 spectrometer, wave numbers are given in cm^{-1}. NMR spectra were measured on FT-NMR spectrometers Bruker AVANCE-500 and Varian UNITY-500 (^{1}H at 500 MHz, ^{13}C at 125.7 MHz) in $CDCl_3$, CD_3COCD_3 and/or C_6D_6. Chemical shifts (^{1}H referenced to TMS, ^{13}C related to the solvent peak $\delta(CDCl_3)$ 77.0 or $\delta(C_6D_6)$ 128.0) are given in ppm (δ-scale), coupling constants (*J*) in Hz. Homonuclear and heteronuclear 2D-NMR experiments (H,H-COSY, H,H-ROESY, H,H-*J*-resolved, H,C-HSQC, and H,C-HMBC) were used for complete structure assignments of proton and carbon signals. Thin-layer chromatography (TLC) was performed on silica gel (ICN Biochemicals) or alumina containing 5% of gypsum. Preparative TLC (PLC) was carried out on 200 × 200 mm plates coated with a 0.7 mm thick layer of the same material; before use, the plates were kept in atmosphere of ammonia for 2 h. Before evaporation on a rotary evaporator in vacuum (bath temperature 50 °C), solutions in organic solvents were dried over anhydrous magnesium sulfate. Whenever solutions of potassium carbonate, potassium hydrogencarbonate or hydrochloric acid were used, their concentration was always 5%. For column chromatography, 60–120 μm silica gel was used. Reversed-phase preparative HPLC was carried out on Separon SGX RPS C-18 (10 μm) in linear water–methanol gradient (40–60% during 20 min).

Epoxidation of Estr-5(10)-ene-3,17-dione (**1**)

A cooled (0 °C) solution of peroxyphthalic acid (7.0 g, 38.4 mmol) in ether (400 ml) was slowly added to a stirred solution of estr-5(10)-ene-3,17-dione (**1**; 10.0 g, 36.7 mmol) in chloroform (50 ml). The mixture was kept for at laboratory temperature 2 h, then washed with the solution of sodium carbonate and water and concentrated in vacuum. The product (a mixture of **2** and **3**, 8.1 g, 78%) melts at 102–144 °C (methanol). TLC (aluminium oxide with 5% of gypsum pre-treated with ammonia, developed with chloroform washed with ammonia): two spots with R_F 0.35 (**2**) and 0.26 (**3**). For ^{1}H and ^{13}C NMR data, see Tables I and II.

10β-Hydroxyestr-4-ene-3,17-dione (**4**) and 5,10β-Dihydroxy-5α-estrane-3,17-dione (**6**)

The mixture of epoxides **2** and **3** (5.0 g, 17.3 mmol) was dissolved in dioxane (100 ml) and the solution was treated with a solution of concentrated sulfuric acid (0.02 ml) in water (20 ml). After 16 h, the acid used was neutralised with the solution of potassium hydrogencarbonate and dioxane was distilled off under reduced pressure. The product was extracted with ethyl acetate, dried, and concentrated in vacuum. Chromatography on a column of silica gel (300 g) in benzene–ether (9:1) with an increasing amount of methanol (0 to 4%) yielded compound **4** (1.3 g, 26%), m.p. 204–205 °C (chloroform–ether); $[\alpha]_D$ +153 (*c* 0.5) (lit.[11] gives 206–207 °C and $[\alpha]_D$ +148). For ^{1}H and ^{13}C NMR data, see Tables I and II. The more polar product was diol **6** (3.1 g, 58%), m.p. 222–228 °C (chloroform–ether); $[\alpha]_D$ +91.8 (*c* 0.25) (lit.[12] gives 223–225 °C and $[\alpha]_D$ +118, lit.[13] gives 240–241 °C and $[\alpha]_D$ +97). For ^{1}H and ^{13}C NMR data, see Tables I and II.

10β-Hydroxyestr-4-ene-3,17-dione (**4**)

A) On silica gel: the mixture of epoxides **2** and **3** (80 mg, 0.28 mmol) was applied on two PLC plates which were kept at room temperature for 2 h. Then they were developed with a solution of methanol (2%) in chloroform. Two major zones were detected by inspection in UV light after spraying the plates with a solution of morin (0.02%). The more lipophilic zone was extracted with ether yielding 10β-alcohol **4** (56 mg, 67%), m.p. 204–205 °C, identical with the sample described above.

B) At increased temperature: a solution of the mixture of compounds **2** and **3** (7.0 g, 24.3 mmol) in acetic acid (250 ml) was heated to reflux for 80 min. The mixture was diluted with chloroform (500 ml) and washed with water (3 × 100 ml) and the sodium carbonate solution. The extract was washed with water, dried, and concentrated in vacuum. Chromatography on silica gel (300 g) in chloroform yielded compound **4** (3.5 g, 50%) and estrone (**7**; 1.2 g, 18%) identical with authentic products.

C) From diol **6**: compound **6** (80 mg) was heated to reflux in acetic acid (10 ml). After 24 h, the solvent was evaporated in vacuum. Remnant of the acetic acid was removed by repeated evaporation with toluene (3 × 10 ml). The remainder was found (^{1}H NMR, TLC in benzene–ether 1:1) to be estrone (**7**) only.

10α-Hydroxyestr-4-ene-3,17-dione (**5**)

The more polar zone from the above PLC yielded 10α-alcohol **5** (28 mg, 33%), m.p. 177–179 and 188–189 °C (acetone–heptane); $[\alpha]_D$ –44.1 (*c* 0.1) (lit.[2] gives 186–188 °C and $[\alpha]_D$ –124). IR: 3595, 3455, 1049 (OH); 1734, 1407 (CH_2–C=O); 1667, 1635 (C=C–CO). FAB MS, *m/z*: 289.2 (M^+), 271.2 ($M^+ - H_2O$). For $C_{18}H_{24}O_3$ (288.4) calculated: 74.97% C, 8.39% H; found: 74.83% C, 8.42% H. For ^{1}H and ^{13}C NMR data, see Tables I and II.

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