DRUG SYNTHESIS METHODS AND MANUFACTURING TECHNOLOGY

A SIMPLE METHOD OF OBTAINING 11-KETO-9β-ESTRA-1,3,5(10)-TRIENES: POTENTIAL REACTANTS FOR THE SYNTHESIS OF STEROIDAL ANTIGESTAGENS

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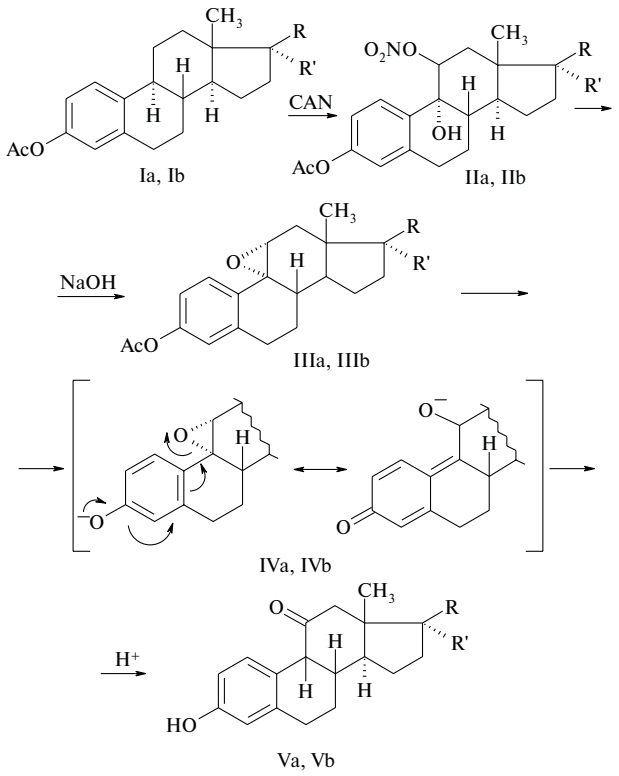
In recent years there have been extensive investigations into the chemistry and biology of 11-aryl-norsteroids known to possess a broad spectrum of physiological properties, in particular, antigestagen activity [1]. These compounds are capable of competing with natural gestagens for binding to receptors of the reproductive organs of females, thus violating normal development of the fetus. In contrast to traditional oral contraceptives, antigestagen preparations can not only prevent but, owing to their abortive action, terminate gestation.

Among the abortive contraceptives, most widely used is the drug mifepristone (RU-486) reproduced in Russia as pencroftone. RU-486 and its analogs are obtained by partial syntheses from estrone. All variants of the synthesis, while differing in the number of stages and/or the way of protecting the 3-keto group, employ only estra-5,9-diene-3,17-dione as the initial compound [2 – 4]. At the same tine, the synthesis of related 11-alkyl-19-norsteroids was frequently performed using 11-ketoestrogens, representing steroids with aromatic ring A [5]. These compounds, albeit considered among possible reactants in the synthesis of 11-aryl-19-norsteroids [6], were never used in practice – apparently because of the lack of sufficiently economic and technologically simple methods for their production.

The known syntheses, for example, of 11-keto-9β-estrone (compounds with the 9β configuration are most stable in the series of 11-ketoestratrienes [7]), include three principal stages: (i) dehydration with 2,3-dichloro-5,6-dicyano-1,4-benzoquinone (DDB), (ii) epoxidation with superacid, and (iii) epoxide conversion into ketone under the action of Lewis acid (BF_3, Et_2O, $LiClO_4$) [7]. All these schemes possess drawbacks related to the difficulties in reproducing the first stage even for simple 17-analogs of estrone; attempts at using relatively cheap and accessible chloranil instead of DDB were unsuccessful [8 – 10].



I – V: R + R′ = O (a); R = β-OAc, R′ = α-C≡CH (b).

Below we describe a two-stage synthesis of 11-keto-9β-estrone (Va) and its 17α-ethinyl analog (Vb). The latter compound was selected because most active gestagens contain

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17α-substituents with C≡C structure (e.g., RU-486 contains a propyl radical).

Taking into account the drawbacks of the aforementioned methods of obtaining 11-keto-9β-estrogens, we decided to use an essentially different pathway for their synthesis. Our synthesis is based on the reactivity of 11-substituted nitrates of 9α,11β-dihydroxysteroids of types IIa and IIb readily obtained through oxidation of estratrienes Ia and Ib with cerium ammonium nitrate (CAN) [11]. A special feature of 11-nitrates IIa and IIb is the easy elimination of HNO_3 under the action of alkalis. The elimination rate exceeds the rate of hydrolysis of the 3-acetoxy group, as evidenced by the synthesis of epoxides IIIa and IIIb at a nearly equimolar amount of the alkali [12].

It could be expected that, with an excess of the alkali, the hydrolysis of 3-acetoxy group in epoxides IIIa and IIIb might lead to phenolates IVa and IVb. However, such compounds are very unstable and several attempts [13 – 15] were unsuccessful. Therefore, rupture of the mobile 9-C–O benzyl bond can lead, via a pathway indicated in the scheme, to stable ketones Va and Vb. Indeed, 11-nitrates IIa and IIb readily convert into ketones Va and Vb within 2.5 – 3 h at room temperature under the action of 4 – 5 mole-eq. NaOH in pyridine. Ketones Va and Vb were isolated with a yield of 82 and 70%, respectively. TLC monitoring showed that intermediate epoxides IIIa and IIIb appear within 30 – 40 min upon mixing the initial nitrates with the alkali.

As is known, nitroesters undergo the reaction of HNO_2 α-elimination under the action of alkalis. An analogous reaction was reported for steroids and well [16]. In the case of 11-nitrates IIa and IIb, this reaction could be expected to yield the corresponding 9α-hydroxy-11-ketosteroids. However, possessing a sample of 9α-hydroxy-11-ketoestrone [17], we established by HPLC that these compounds did not appear in the reaction. Thus, a preferred reaction pathway for the nitrates of vicinal diols II is the β-elimination of nitric acid rather than the β-elimination process.

Ketone Va appears as a crystalline, high-melting substance with physicochemical characteristics corresponding to those reported in [11]. Ketone Vb was isolated in the form of a foam, but the spectroscopic data confirmed the proposed 9β-11-ketone structure. In the ^{1}H NMR spectrum of ketone Vb (as well as in that of ketone Va), the signal from the equatorial 9-H proton is manifested as a broad doublet. The spin – spin coupling constant is 3.5 Hz for Va and 4.3 Hz for Vb, which corresponds to the electron-acceptor interaction with 8-H proton. Both ketones are characterized by a low-field position of the signal from 1-H proton, caused by the descreening effect of the 11-keto group. This group is manifested in the IR spectra of ketones Va and Vb as an intense absorption band at 1700 cm^{-1} (Va) and 1680 cm^{-1} (Vb).

It should be noted that the proposed synthesis of 11-keto-9β-estra-1,3,5(10)-trienes Va and Vb differs from the known processes in that the number of stages is less, the procedure is simpler, and no expensive reactants (such as DDB) are involved.

EXPERIMENTAL PART

The melting points were determined using a Boetius heating table. The optical rotation was studied with a Polamat A spectropolarimeter. The UV spectra were recorded on a Specord UV-VIS spectrophotometer as ethanol solutions; the IR spectra were measured on a Specord 75-IR spectrophotometer in KBr pellets. The ^{1}H NMR spectra were measured with Bruker Model 360 and Tesla BS-587A 80-MHz instruments using $CDCl_3$ as the solvent and TMS as the internal standard (unless otherwise indicated). The HPLC measurements were performed on a Milikhrom-1 system equipped with a UV detector tuned to 280 nm. A 80 × 2 mm Silasorb C18 column was eluted with an acetonitrile – water (7 : 3) mixture; peak retention times (RRT) were determined in minutes. The compositions of reaction mixtures and the purity of products were monitored by TLC on Silufol UV-254 plates. The data of elemental analyses performed for compounds IIb and IIc coincided with the results of analytical calculations.

General method for obtaining nitrates IIa and IIb. To a solution of 1 mmole of compound Ia or Ib in 15 ml of acetic acid was added dropwise a solution of 2.52 g (4.6 mmole) of CAN in 2 ml of water. The orange solution was stirred for 2 h at room temperature, diluted by half with water, and extracted with chloroform or ethyl acetate. The extract was sequentially washed with saturated solutions of $NaHCO_3$ and NaCl and dried over $MgSO_4$. The target products were isolated by chromatography on silica gel (10 g sorbent per gram mixture) eluted with an ethyl acetate – hexane (7 : 3) mixture.

3,9α-Dihydroxy-11β-nitroxyestra-1,3,5(10)-trien-17-one-3-acetate (IIa). Yield, 56%; m.p., 174 – 176°C; $[\alpha]_D$, +103 ± 4° (*c*, 0.86; chloroform) (reported [13]: m.p., 176.5 – 178°C; $[\alpha]_D$, +100°).

3,9α,17β-trihydroxy-11β-nitroxy-17α-ethinylestra-1,3,5(10)-triene-3,17-diacetate (IIb). Yield, 44%; m.p., 172 – 174°C (methanol); $[\alpha]_D$, –22 ± 2° (*c*, 0.95; chloroform); UV spectrum, λ_{max}, nm (log ε): 266 (2.75), 274 (2.70); IR spectrum (ν_{max}, cm^{-1}): 3430 (OH), 3250 (C≡CH), 1747 (3-$OCOCH_3$), 1735 (17-$OCOCH_3$), 1630, 1500 (C=C arom.), 1240, 1030 (C–O); ^{1}H NMR spectrum (δ, ppm): 1.03 (s, 3H, 18-CH_3), 2.04 (s, 3H, 17-$OCOCH_3$), 2.27 (s, 3H, 3-$OCOCH_3$), 2.7 (s, 1H, C≡CH), 5.85 (t, 1H, J 3.0 Hz, 11-H), 6.88 (bs, 1H, 4-H), 6.92 (d, 1H, J 9.0 Hz, 2-H), 7.30 (d, 1H, J 9.0 Hz, 1-H).

3-Hydroxy-9β-estra-1,3,5(10)-triene-11,17-dione-11-keto-9β-estrone (Va). To a solution of 4.38 g (11.26 mmole) of nitrate IIa in 80 ml of pyridine in an argon atmosphere was added 8 ml of a 20% aqueous NaOH solution (40 mmole) and the mixture was stirred at room temperature, whereby the solution becomes yellow (15 min), then turns violet and dark-violet (1.5 h), and $NaNO_3$ is precipitated. After a 3-h stirring, the reaction mixture was diluted by half with water, acidified (on cooling) to pH 4 – 5 with a 10% aqueous HCl

solution, and extracted with ethyl acetate. The extract was sequentially washed with a 10% aqueous HCl solution and a saturated NaCl solution and dried over $MgSO_4$. The ethyl acetate was evaporated, the residue was dissolved in chloroform, and the chloroform solution was filtered through silica gel (40 g), after which ketone Va was isolated; yield, 2.6 g (82%); m.p., 199 – 204°C (methanol); $[\alpha]_D$, +259 ± 15° (*c*, 0.84; dioxane) (reported [11]: m.p., 194 – 198°C and 204 – 207°C $[\alpha]_D$, +244°; RRT, 4.8 min); IR spectrum (ν_{max}, cm^{-1}): 3320 (OH), 1725 (17-CO), 1700 (11-CO), 1610, 1450 (C=C arom.), 1230 (C–O); ^1H NMR spectrum in C_5D_5N (δ, ppm): 0.85 (s, 3H, 18-CH_3), 3.75 (bd, 1H, J 3.5 Hz, 9-H), 6.87 – 7.05 (m, 2H, 2-H, 4-H), 7.27 (d, 1H, J 9.0 Hz, 1-H).

3,17β-Dihydroxy-17α-ethinyl-9β-estra-1,3,5(10)-trien-11-one-17-acetate (Vb). Compound Vb was obtained with a yield of 70% using a procedure analogous to that described for compound Va. Analytical sample in the form of a colorless foam was isolated by preparative TLC on a 20 × 20 cm Silufol UV-254 plate eluted in an ethyl acetate – hexane (1 : 1) mixture; RRT, 3.8 min; IR spectrum (ν_{max}, cm^{-1}): 3360 – 3240 (OH), 3260 (C≡CH), 1710 – 1725 (CO in $OCOCH_3$), 1680 (CO), 1610, 1575, 1495 (C=C arom.). ^1H NMR spectrum in C_5D_5N (δ, ppm): 0.98 (s, 3H, 18-CH_3), 2.03 (s, 3H, $OCOCH_3$), 2.39 (s, 1H, C≡CH), 3.62 (bs, 1H, J 4.3 Hz, 9-H), 5.1 (bs, 1H, OH), 6.6 (d, 1H, J 9 Hz, 2-H), 6.62 (bs, 1H, 4-H), 6.85 (d, 1H, J 9 Hz, 1-H).

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