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INTERACTION OF 3-β-CHLORO-Δ^5-STEROIDS WITH TRIFLUOROPERACETIC ACID

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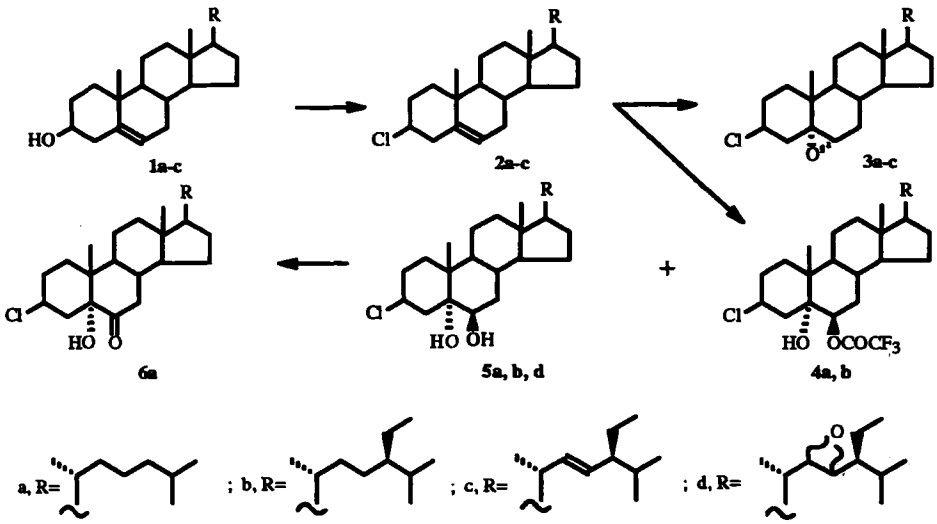
*It has been established that the reaction of 3β-chloro-Δ^5-steroids with trifluoroperacetic acid forms not only the 5α,6-peroxides (**3a,d**), but also the 5α,6β-diols (**5a,b,d**) and their 6-trifluoroacetates.*

The Prilezhaev [Prileschajew] oxidation of a double bond in steroids with per-acids is a well-studied reaction [1]. This method is widely used for the synthesis of 5,6-epoxides, which are important intermediates in the production of ecdysteroids [2]. For epoxidation it has been proposed to use perbenzoic, *m*-chloroperbenzoic, monoperphthalic [1], *o*-sulfoperbenzoic [3], *p*-methoxycarbonylperbenzoic [4], and pentafluoroperbenzoic [5] acids. It must also be mentioned that when performic acid, obtained directly in the reaction mixture from hydrogen peroxide and formic acid, was used for the oxidation of Δ^5-steroids, the 5,6-epoxides formed underwent immediate transformation into the corresponding 5α,6β-diols [6].

By the epoxidation of 3β-chloro-Δ^5-stigmastanes (**2b,c**) with *m*-chloroperbenzoic acid we have previously [7] obtained the 5α,6α-epoxides (**3b,c**), which were then used for the synthesis of 5α-hydroxy derivatives of brassinosteroids. With the aim of expanding the possibilities of the synthesis 5α,6α-epoxides of the type of (**3a—d**), we have investigated the products of the oxidation of compounds (**2a—c**) in trifluoroperacetic acid obtained directly through the reaction of trifluoroacetic anhydride and hydrogen peroxide. This reagent is commonly used for the epoxidation of compounds with nucleophilic double bonds [8].



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We obtained the initial 3β-chloro-Δ^5-steroids (**2a—c**) by procedures developed previously [7, 9] for the reactions of cholesterol (**1a**), β-sitosterol (**1b**), and stigmasterol (**1c**), respectively, with thionyl chloride. We found that the interaction of the 3β-chloro-Δ^5-steroids (**2a—c**) with trifluoroperacetic acid in chloroform or methylene chloride takes place in fairly complex fashion with the formation of at least three products in each case. Thus, on the oxidation of 3β-chlorocholest-5-ene (**2a**) by this reagent in chloroform, followed by chromatographic purification, we succeeded in isolating a mixture of the 5α,6α-epoxide (**3a**) with the 6-trifluoroacetate of the 5α,6β-diol (**4a**), and the pure 5α,6β-diol (**5a**). The structure of the latter followed unambiguously from its spectra. In particular, its IR spectrum had an intense band at 3440 cm^{-1} corresponding to the stretching vibrations of hydroxy groups.

The ^1H NMR spectrum of the diol (**5a**) lacked the signal of the H-6 vinyl proton, while the signal of the methine proton geminal to the chlorine atom, H-3α, appeared at 4.36 ppm. From the half-width of the H-3α signal (W/2 = 24 Hz) it was possible to deduce its axial orientation and, since the configuration at C-3 did not change in the course of the reaction, the *trans*-linkage of rings *A* and *B*. The observed downfield shift of the H-3 signal in comparison with its position in the spectrum of the initial compound (**2a**) (δ 3.8 ppm) served as proof of the presence in compound (**5a**) of a 5α-hydroxy group in a 1,3-diaxial position with respect to H-3α.

Moreover, the spectrum of steroid (**5a**) included at 3.54 ppm a multiplet of the proton geminal to the 6-hydroxy group. The half-width of this signal (W/2=8Hz) related to equatorial orientation of H-6 atom and therefore to axial or β-orientation of 6-hydroxy group. The fact that compound (**5a**) had a 6β-hydroxy group was also confirmed by a downfield shift to 1.21 ppm of the signal of the angular 19-methyl group in the 1,3-diaxial position with respect to this hydroxy group.

In order to obtain additional proofs of its structure, compound (**5a**) was subjected to Jones oxidation with chromic acid in acetone. The product of this reaction proved to be completely identical with 3β-chloro-5α-hydroxy-6-ketone (**6a**) that we synthesized by a method developed previously [7] — the epoxidation of the 3β-chlorocholest-5-ene (**2a**) with *m*-chloroperbenzoic acid, followed by the Jones oxidation of the resulting 3β-chloro-5α,6α-epoxide (**3a**).

The structures of compounds (**3a**) and (**4a**), which we were unable to separate, were shown by an analysis of spectra of their mixture. Thus, the ^1H NMR spectrum of this mixture had a characteristic doublet (δ 2.82 ppm, J = 4.8 Hz) corresponding to the absorption of the H-6β proton geminal to the 5α,6α-epoxy group. The same signal was present in the spectrum of the pure 5α,6α-epoxide (**3a**) obtained by the oxidation of compound (**2a**) with *m*-chloroperbenzoic acid. In addition, in the spectrum of the mixture of compounds (**3a**) and (**4a**) there was a signal of a methine proton with δ 4.10 ppm in the form of a multiplet with the half-width W/2 = 21 Hz. Precisely the same signal, corresponding to the resonance absorption of H-3α, was present in the spectrum of the pure epoxide (**3a**). The fact that the mixture also contained the 6-trifluoroacetate of the 3β-chloro-5α,6β-diol (**4a**) was shown by the presence in the ^1H NMR spectrum of the signals of H-3α (δ 4.30 ppm, W/2 = 25 Hz) and H-6α (δ 4.82 ppm, W/2 = 26 Hz). Here, attention is attracted by the considerable downfield shift of H-6α signal in comparison with its position in the spectrum of the diol (**5a**). Such a shift is observed in the case of esterification of a hydroxy group.

The structure of the second component of the mixture was likewise shown by the presence in the IR spectrum of a band at 1790 cm^{-1}, the position of which is extremely characteristic for trifluoroacetates. The results of hydrolysis of a mixture of compounds (**3**) and (**4a**) under the action of an aqueous solution of sodium hydroxide in a mixture of ethanol and tetrahydrofuran confirmed the conclusions about their structures made on the basis of spectral analysis. Thus, in addition to the 5α,6α-epoxide (**3a**) that had not reacted, we isolated the 5α,6β-diol (**5a**). The structures of these substances were shown by comparison with authentic specimens.

The trifluoroperacetic acid oxidation of 3β-chlorostigmastene (**2b**), obtained from β-sitosterol, took place similarly. By the chromatographic separation of the mixture of reaction products we succeeded in isolating the pure 5α,6α-epoxide (**3b**), a mixture of it with the 6-trifluoroacetate (**4b**), and the 5α,6β-diol (**5b**). Alkaline hydrolysis of the mixture of (**3b**) and (**4b**) under the action of an aqueous solution of sodium hydroxide in a mixture of ethanol and tetrahydrofuran enabled us to obtain additional amounts of the 5α,6α-epoxide (**3b**) and the 5α,6β-diol (**5b**). The overall yields of these compounds were 15 and 53%, respectively. The structures of compounds (**3b**) and (**5b**) followed unambiguously from their spectra.

After 3-chlorostigmasterol (**2c**), obtained from stigmasterol (**1c**), had been oxidized with trifluoroperacetic acid, we subjected the reaction products to alkaline hydrolysis with a solution of sodium hydroxide immediately, without their separation. In this way it was possible to obtain the 5α,6β-diol (**5d**) with a yield of 65.4%. The structure of compound (**5d**) was also unambiguously confirmed by its IR and ^1H NMR spectra. Important for determining the structure of the side-chain was the absence of the H-22 and H-23 vinyl protons from the ^1H NMR spectrum of the substance under discussion. At the same time,

characteristic signals of H-22 and H-23 methine protons geminal to a 22,23-epoxy group appeared in the spectrum at 2.50 and 2.75 ppm.

Judging from its spectra, compound (**5d**) was a mixture of (22R,23R)- and (22S,23S)- epimers. It must be mentioned that such epimers are also commonly formed on the *m*-chloroperbenzoic acid epoxidation of a 22(23)-double bond in the stigmastane series.

Thus, we have established that, with a careful choice of reaction conditions, the 5α,6β-diols (**5a, b,** and **d**) can be obtained in fairly high yield by the oxidation of compounds (**2a—c**) with trifluoroperacetic acid. However, in this case, apparently, the efficiency of trifluoroperacetic acid is always inferior to that of performic acid.

EXPERIMENTAL

Melting points were determined on a Kofler block. IR spectra were recorded on a UR-20 instrument in the range of 700—3600 cm^{-1} in KBr tablets. ^1H NMR spectra of solutions in deuterochloroform were obtained on Bruker WM-360 and Bruker AC-200 instruments with working frequencies of 360 and 200 MHz. Chemical shifts are given relative to TMS as internal standard. For column chromatography we used Lachema (Czech Republic) type L40/100 silica gel and Reanal (Hungary) alumina (activity grade II). The course of the reactions was monitored and the purity of the compounds obtained was checked with the aid of Kavalier (Czech Republic) Silufol plates.

Oxidation of 3β-Chlorocholest-5-ene (2a) with Trifluoroperacetic Acid. In portions, with stirring and cooling, 25 ml of 30% hydrogen peroxide solution was added to a solution of 22.8 ml of trifluoroacetic anhydride in 50 ml of chloroform. With continued stirring and cooling, 5.0 g of 3β-chlorocholest-5-ene (**2a**) (obtained from cholesterol (**1a**) by the procedure of [9]) in 20 ml of chloroform was added to the emulsion so obtained.

The reaction mixture was stirred at room temperature for 10 min and was then filtered through a layer of alumina, and the filtrate was evaporated in vacuum. The residue, which, to judge from TLC, contained the initial substance, was dissolved in 10 ml of chloroform and reoxidized with a solution of trifluoroperacetic acid for 30 min. The reaction mixture was filtered through a layer of alumina, the filtrate was evaporated in vacuum, and the residue was chromatographed on a column of silica gel.

Elution with benzene—tetrahydrofuran (5:1) yielded 2.6 g of a main fraction, which was rechromatographed with elution by hexane. This gave two fractions. Fraction 1 consisted of 1.39 g of a mixture of 3β-chloro-5,6α-epoxy-5α-cholestane (**3a**) and 3β-chloro-5α-cholestan-5,6β-diol 6-trifluoroacetate (**4a**). Fraction 2 was 0.41 g (7.6%) of 3β-chloro-5α-cholestane-5,6β-diol (**5a**). mp 99—102°C (hexane).

IR spectrum (KBr, ν, cm^{-1}): 3440 (OH). ^1H NMR spectrum (δ, ppm.): 0.68 (s, 19-Me), 0.86 (d, J=7 Hz, 26/27-Me), 0.90 (d, J=7 Hz, 21-Me), 1.21 (s, 19-Me), 2.46 (t, J=13 Hz, H-4β), 3.54 (m, W/2=8 Hz, H-6α), 4.36 (m, W/2=24 Hz, H-3α).

With stirring, 4 ml of a 5% aqueous solution of sodium hydroxide was added to a solution of 0.41 g of fraction 11 in a mixture of 30 ml of ethanol and 10 ml of tetrahydrofuran. The reaction mixture was stirred at room temperature for 2 h and was then neutralized with dilute (1:1) acid to pH 4—5, and, after the addition of 50 ml of water, it was extracted with 50 ml of methylene chloride. The extract was washed with water and evaporated in vacuum. The residue was chromatographed on a column of silica gel, with elution by benzene—tetrahydrofuran (20:1). This yielded 0.41 g (7.9%) of 3β-chloro-5,6α-epoxy-5α-cholestane (**3a**).

Epoxide (**3a**) had a ^1H NMR spectrum identical with that of the substance obtained by the epoxidation of chlorocholestane (**2a**) with *m*-chloroperbenzoic aid. Further elution gave 0.46 g (8.5%) of the diol (**5a**), mp 107—116°C (hexane). The total yield of the diol (**6a**) was 16.1%.

3β-Chloro-5,6α-epoxy-5α-cholestane (3a). To a solution of 18.23 g of 3β-chlorocholest-5-ene (**2a**) in 100 ml of chloroform was added 8.93 g of *m*-chloroperbenzoic acid (content of the main substance 85%). The reaction mixture was kept at room temperature for 27.5 h and was then neutralized with 160 ml of a saturated solution of sodium bicarbonate with stirring on a magnetic stirrer for 35 min. The organic layer was separated from the aqueous layer, washed with 50 ml of water, and evaporated in vacuum. The residue was chromatographed on a column of alumina, with elution by pentane—ether (5:1). This gave 17.51 g (92%) of the epoxide (**3**), mp 78—83°C (pentane).

^1H NMR spectrum (δ, ppm): 0.61 (s, 18-Me), 0.86 (t, J=7.2 Hz, 26-Me), 0.87 (t, J=7.2 Hz, 27-Me), 0.89 (t, J=7.2 Hz, 21-Me), 1.09 (s, 19-Me), 2.28 (t, J=9.6 Hz, H-4β), 2.82 (d, J=4.8 Hz, H-6β), 4.10 (tt, J_1-13 Hz, J_2=5 Hz, H-3α).

3β-Chloro-5-hydroxy-5α-cholestan-6-one (6a). A. A solution of 16.69 g of the epoxide (**3a**) in 700 ml of acetone was treated with 24 ml of 8 N chromic acid, and the mixture was kept at room temperature for 25 min. The excess of oxidant was neutralized with propan-2-ol. The mixture was filtered, and the filtrate was diluted with water and extracted with chloroform. The organic extract was evaporated in vacuum and the residue was chromatographed on a column of alumina with elution by petroleum ether—chloroform (1:1). This yielded 3.99 g (23%) of the 5-hydroxy-6-ketone (**6a**), mp 180—186°C (acetone—petroleum ether).

IR spectrum (KBr, ν, cm^{-1}): 3515 (OH), 1710 (C=O).

^1H NMR spectrum (δ, ppm.): 0.64 (s, 18-Me), 0.82 (s, 19-Me), 0.82 (d, J=6 Hz, 26-Me), 0.85 (d, J=6 Hz, 27-Me), 0.90 (d, J=6 Hz, 21-Me), 2.68 (t, J=12 Hz, H-4β), 4.20 (m, W/2=24 Hz, H-3α).

B. With stirring at 15°C, 0.8 ml of 8 N chromic acid was added to a solution of 0.80 g of the diol (**5a**) in 10 ml of acetone. The reaction mixture was stirred at 15°C for 10 min, and the excess of oxidant was eliminated by the addition of 1.0 ml of isopropanol. After dilution with 10 ml of water the reaction product was extracted with dichloroethane, and the extract was washed with water and evaporated in vacuum, giving 0.075 g (95%) of ketone (**6a**), mp 170—176°C. Compound (**6a**) had the same IR and ^1H NMR spectra as an authentic specimen obtained by method A.

Oxidation of (24R)-3β-Chlorostigmast-5-ene (2b) with Trifluoroperacetic Acid. In portions, with stirring and cooling, 13 ml of 30% hydrogen peroxide was added to a solution of 23 ml of trifluoroacetic anhydride in 40 ml of chloroform. A solution of 4.3 g of (24R)-3β-chlorostigmast-5-ene (**2b**) (obtained from β-sitosterol by the procedure of [9]) in 15 ml of chloroform was added to the emulsion so formed. The reaction mixture was stirred at room temperature for 30 min and was then filtered through a layer of alumina. The filtrate was evaporated in vacuum and the residue was chromatographed on a column of silica gel. On elution with benzene—tetrahydrofuran (20:1), three fractions were obtained.

Fraction 1 — 0.174 g (4%) of (24R)-3β-chloro-5,6α-epoxy-5α-stigmastane (**3b**). The substance had a ^1H NMR spectrum identical with that of an authentic specimen obtained earlier by the method of [7]. Fraction 2 — 2.24 g of a mixture of (24R)-3β-chloro-5,6α-epoxy-5α-stigmastane (**3b**) and (24R)-3β-chloro-5α-stigmastane-5,6β-diol 6-trifluoroacetate (**4b**). Fraction 3 — 0.47 g (11%) of (24R)-3β-chloro-5α-stigmastane-5,6β-diol (**5b**), mp 128—135°C (hexane—ether).

IR spectrum (KBr, ν, cm^{-1}): 3430 (OH). ^1H NMR spectrum (δ, ppm.): 0.68 (s, 18-Me), 1.20 (s, 19-Me), 2.46 (t, J=12.5 Hz, H-4β), 3.54 (m, W/2=8 Hz, H-6α), 4.36 (m, W/2=24 Hz, H-3β).

A solution of 2.24 g of fraction 2 in 30 ml of ethanol and 10 ml of tetrahydrofuran was treated with 4 ml of a 5% solution of sodium hydroxide. The reaction mixture was stirred at room temperature for 2 h and was then neutralized with dilute hydrochloric acid (1:1) to pH 4—5, and, after the addition of 50 ml of water, it was extracted with 50 ml of methylene chloride. The extract was washed with water and evaporated in vacuum. The residue was chromatographed on a column of silica gel, with elution by hexane—tetrahydrofuran (30:1). Two fractions were obtained. Fraction 1 — 0.49 g (11%) of the epoxide (**4b**). Fraction 2 — 1.81 g (42%) of the diol (**5b**), mp 127—141°C (hexane).

Oxidation of (24S)-3β-Chlorostigmasta-2,22-diene (3c) with Trifluoroperacetic Acid. In portions, with stirring and cooling, 8.5 ml of a 30% solution of hydrogen peroxide was added to a solution of 15 ml of trifluoroacetic anhydride in 30 ml of methylene chloride. With stirring, a solution of 3.0 g of (24S)-3β-chlorostigmasta-2,22-diene (**3c**) (obtained from stigmasterol (**1c**) by the procedure of [7]) in 30 ml of methylene chloride was added to the emulsion so formed. The reaction mixture was stirred at room temperature for 30 min and was then filtered through a layer of alumina. The filtrate was evaporated in vacuum, the residue (2.3 g) was dissolved in a mixture of 30 ml of ethanol and 10 ml of tetrahydrofuran, and 4 ml of a 5% solution of sodium hydroxide was added.

The reaction mixture was stirred at room temperature for 1.5 h and was then neutralized with dilute (1:1) hydrochloric acid to pH 4—5, treated with 50 ml of water, and extracted with 50 ml of methylene chloride. The extract was washed with water and evaporated in vacuum. The residue was chromatographed on a column of silica gel with elution by benzene—tetrahydrofuran (25:1). This gave 2.19 g (65.4%) of (24S)-3β-chloro-22,23-epoxy-5α-stigmastane-5,6β-diol (**5d**), mp 214—215°C (hexane-tetrahydrofuran).

IR spectrum (KBr, ν, cm^{-1}): 3480 (OH). ^1H NMR spectrum (δ, ppm.): 0.65 (s, 18-Me), 1.21 (s, 19-Me), 2.48 (t, J=13 Hz, H-4β), 2.50 m and 2.75 dd (J_1-7.0 Hz, J_2=2.5 Hz, H-22 and H-23), 3.55 (m, W/2=7 Hz, H-6α), 4.38 (m, W/2=25 Hz, H-3α).

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