STEROIDS XXXIX. TOWARDS A METHOD OF SPLITTING SOLASODINE TO $\Delta^{5,16}$-PREGNADIEN-3β-OL-20-ONE

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The industrial method of preparing dehydropregnenolone (Va) from solasodine (I) [1] which was developed by us includes a four-stage process. It operates, without isolation of intermediates, in an overall yield of 56-60% [2,3]; solasodine (I) → solasodine O,N-diacetate (II) → 26-acetylamino-$\Delta^{5,20(22)}$-furostadien-3β-ol acetate (III), ψ-solasodine O,N-diacetate) → 16β-(δ-acetylamino-γ-methylvaleryloxy)-Δ^5-pregnen-3β-ol-20-one (IV, oxidation product) → dehydropregnenolone acetate (Va, DPA). The introduction of the method indicated into industry and the necessity of developing large tonnage production of dehydro-pregnenolone acetate for the synthesis of corticosteroids and sex hormones required a detailed study of each stage of the process to expose side reactions taking place in the event of a variation from the technological routine.

Yields have been established for the individual stages of the route for converting analytically pure products at each stage (II, III, and IV) into DPA (Va). Average yields amounted to:

(I) → (V) 56%, acetylation of (I) → (II) was 92%
(II) → (V) 61%, isomerization of (II) → (III) was 93%
(III) → (V) 66%, oxidation of (III) → (IV) was 70%
(IV) → (V) 95%, cleavage of (IV) → (V) was 95%.

The results of a study of the acetylation of solasodine are cited below. The presence in the molecule (I) of tetrahydrofuran and piperidine rings mean that the yield and constitution of compounds formed on acetylation vary sharply on changing the reaction conditions [2,4-6]. The acetylation of (I) under our suggested conditions was carried out with acetic anhydride in boiling toluene in the presence of calcium oxide with azeotropic distillation of water [2].

26-Acetylamino-Δ^5-furosten-3β, 22-diol 3-acetate (VIa) (~ 1.5%), identified as its methyl ether (VIb), and 26-acetylamino-Δ^5-cholestene-3β, 16β-diol-22-one 3,16-diacetate (VII) (~2%) were isolated by chromatographic separation on silica gel as byproducts of acetylation. Both compounds have been described previously [5]; their constitutions were confirmed additionally by NMR spectra. The acetylation of (I) without azeotropic distillation of water led to a reduction in the yield of DPA to 48-50%. A similar reduction in the yield of DPA was observed on acetylating solasodine (I), the water of crystallization of which (solasodine contains 1 mole or 4% water) had been removed first by azeotropic distillation. On chromatographic separation of the acetylation products an increased amount of (VIa) and (VII) was found (~ 7%) in both cases.

The formation of (VIa) may be explained by acid hydrolysis of solasodine O,N-diacetate (II) [7] (the water distilled off on acetylation contained up to 20% acetic acid). [Scheme on following page].

Solasodine O,O,N-triacetate (X) is a possible precursor of (VII), since it is known that acid hydrolysis of the former leads to the formation of (VII) [5]. Indirect proof of this follows from the fact that ~1.5% (X) and a significant amount (~ 16%) of (VII) were found in the reaction products on acetylating (I) with 80% acetic anhydride by the method we have suggested. (X) was obtained on boiling (I) with acetic anhydride [5]

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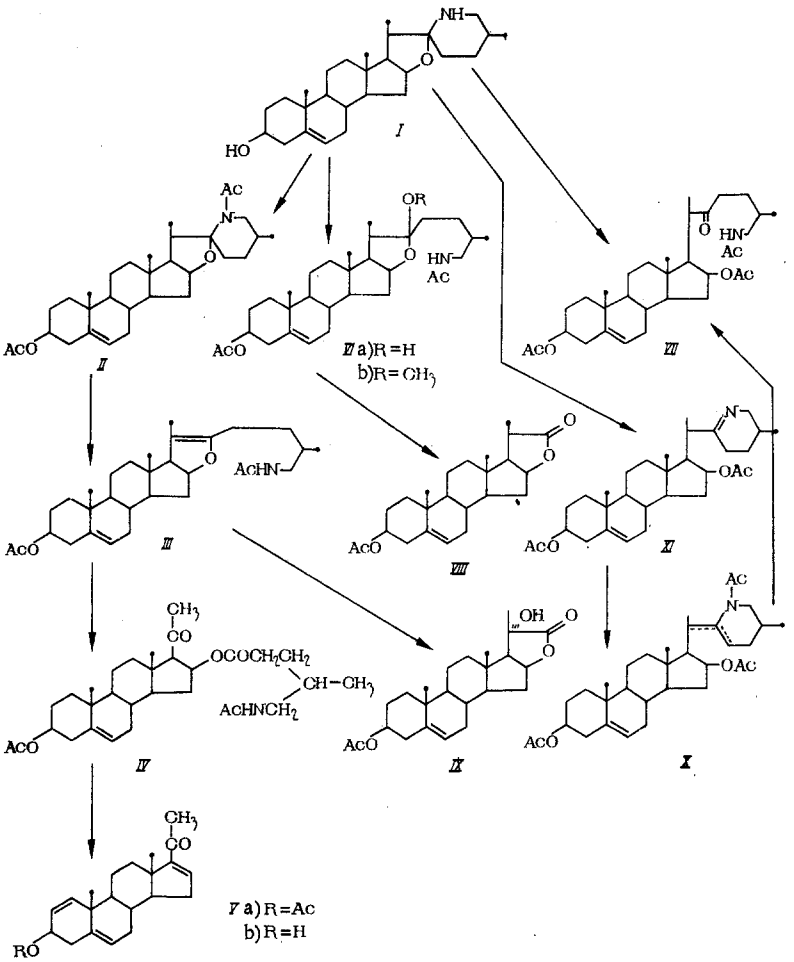

and also by acetylating solasodine-B 3,16-diacetate (XI) [5]. (XI) was formed on acetylating (I) with a mixture of acetic anhydride and acetic acid in the presence of zinc chloride [8] or toluene-p-sulfonic acid [9].* We have shown that acetylating (XI) with acetic anhydride in toluene in the presence of calcium oxide gave (X).

Transformations of (VIa) and (VII) in the subsequent stages of splitting towards DPA have been studied. In the isomerization stage, i.e., boiling in glacial acetic acid for 0.5 h, (VIa) was completely converted into the O,N-diacetate of ψ-solasodine (III) [7] with its subsequent conversion in turn to DPA in 64% yield, i.e., practically the same yield as (II) is converted into DPA. In this case if dehydration does not proceed to completion, then (VIa) forms the (22 → 16) lactone of 3β-acetoxy-16-hydroxy-Δ^5-bisnorcholenic acid (VIII) under our proposed oxidation conditions. It has been established that oxidation of analytically pure (VIa) gives the lactone in 43.5% yield. Contamination by lactone (VIII) in technical DPA leads to a reduction of melting point and increased loss of DPA on crystallization.

Ketone (VII), under the further transformation conditions, is practically unchanged. Analytically pure (VII) having passed the stages of isomerization, oxidation, and cleavage, was isolated unchanged in 80% yield. Contamination by (VII) in technical DPA reduced its melting point; if the content of (VII) was not greater than 10% it remained in the mother liquors when DPA was recrystallized from ethyl alcohol and did not increase the normal loss (8-10%) of DPA on crystallization. If the content of (VII) was more than 10%, then a second crystallization of DPA was required.

Thus, a reduction of the aqueous acetic acid content in the reaction mass on acetylation for various reasons (acetic anhydride of less than 98% content, weak boiling of the reaction solution and consequent poor separation of aqueous acetic acid in the azeotropic mixture, running in acetic anhydride in the cold) leads to a fall in the yield of DPA.

*(XI) was isolated previously from the acetic acid mother liquor as an unidentified compound of mp 198-199°.

TABLE 1. Chromatographic Mobilities (R_f) of Compounds (I)-(XI)

Compound	System 1: chloroform – acetone (9:1)	System 2: benzene – ethyl acetate (8:2)	System 3: cyclohexane – acetone (7:3)	System 4: cyclohexane – ethyl acetate (8:2)
I	0,14	0,05	0,45	0,07
II	0,72	0,36	0,49	0,24
III	0,29	0,04	0,28	0
IV	0,15	0	0,13	0
Va	0,77	0,70	0,55	0,50
Vb	0,49	0,34	0,44	0,21
VIa	0,06	0	0,15	0
VIa	0,26	0,02	0,30	0
VII	0,16	0	0,15	0
VIII	0,75	0,59	0,51	0,39
IX	0,49	0,34	0,37	0,15
X	0,67	0,30	0,44	0,13
XI	0,33	0,04	0,51	0,08

Note: Chromatography was carried out on plates of size 18 × 24 cm. Length of run of solvent was 20 cm.

We have studied the mother liquors from the recrystallization of technical DPA. The alcoholic mother liquor from DPA of mp 166-170° obtained under the normal process conditions did not contain (VIII) or (VII) [(VII) was detected chromatographically in the acetic anhydride mother liquor of DPA]. The absence of (VIII) indicated that under normal conditions (VIa) was completely dehydrated to (III). The mother liquors contained the hitherto undescribed (22 → 16) lactone of 3β-acetoxy-16, 20ξ-dihydroxy-Δ^5-bisnorcholenic acid (IX), the structure of which was established by comparison of the spectroscopic data for (VIII) and (IX). Thus in the IR spectrum of (IX), together with frequencies of 1780 cm^{-1} (CO lactone) and 1735 cm^{-1} (CO ester), which are also characteristic for (VIII), there is an absorption of frequency 3400 cm^{-1}, which must be attributed to a hydroxyl group since there is no nitrogen in the (IX) molecule. On comparing the mass spectra of (VIII) and (IX) it was discovered that both compounds had a common type of fragmentation. Both spectra had no peak for the molecular ion but had M-AcOH of m/e 326 for (VIII) and 342 for (IX) and M-AcOH-CH_3 of m/e 311 for (VIII) and 327 for (IX). The shift in m/e of 16 for (IX) compared to (VIII) indicated the presence in it of a hydroxyl group. The position of the hydroxyl group in the (IX) molecule was established with the aid of NMR spectra. The methyl group at C_{20} in (VIII) had a clearly expressed doublet at 1.13 ppm, due to splitting with the proton at C_{20}. In (IX) this signal had disappeared, but a singlet signal appeared at 1.54 ppm, which corresponds to the signal of a methyl group at a carbon atom linked with an oxygen atom. (IX) is the product of incomplete oxidation of the $C_{20}-C_{22}$ double bond in (III), since (IX) was also isolated on oxidation of analytically pure (III) under normal process conditions.

On deviating from the normal conditions for carrying out the process a reduction in the melting point of technical DPA was observed and additional contaminants appeared in the alcoholic mother liquor, viz., (VII), (VIII), and Δ^5-pregnadien-3β-ol-20-one (Vb), the appearance of which is due to saponification of the acetyl group in position 3.

The study of the conditions of forming byproducts at the stage of cleavage of solasodine and their transformations in the course of subsequent reactions leading to DPA has made possible a series of recommendations for conducting the technological process at the stages of acetylation, isomerization, and cleavage.

EXPERIMENTAL

IR spectra were taken on a UR-10 instrument and NMR spectra on an INM-4H-100 instrument in deuterochloroform with tetramethylsilane as internal standard; mass spectra were determined on an MX-1303 instrument provided with a system for introducing specimens directly into the ion source at an electron energy of 70 eV and 140-145°. Chromatography was carried out on plates in thin layers of silica gel type KSK with gypsum as binder using phosphomolybdic acid at 100° for 5 min for visualization (see Table 1). Preparative chromatography was effected on the same type of silica gel. On describing chromatographic resolutions on columns, fractions containing mixtures of compounds are not indicated if they were not subjected to further treatment. Solutions intended for evaporation in vacuum were dried over anhydrous sodium sulfate. The identity of the compounds isolated with known specimens was established on the basis of a lack of mp depression in mixing tests and comparison of IR spectra and chromatographic mobilities. Solasodine of 93% content was used (allowing for water of crystallization).

Acetylation of Solasodine (I). A. Standard conditions. After acetylation of 10 g solasodine (I) the toluene solution was evaporated to dryness, the oily residue (12 g) dissolved in acetone, the solid filtered off and recrystallized from acetone. Five grams (II) was obtained with mp 158-160° [2]. After evaporation of the mother liquor 7 g oily residue was chromatographed on 85 g silica gel. A mixture of methylene chloride and ethyl acetate (7:3) washed off 4.96 g (II), which, after recrystallization from acetone, had mp 159-161°. A mixture of methylene chloride and ethyl acetate (1:1) washed off 0.27 g (2.2%) (VII) of mp 176-177° (from ethyl alcohol) [5]. NMR spectrum: δ 0.83, 0.99, 1.09 (CH_3 group), 1.91, 1.95, 1.99 (singlet

CH_3CO ester, amide), 2.31, 2.97 (multiplet, $CH_2C{=}O$, $CH_2C{-}NH$), 4.55, 4.9 (multiplet 3αH, 16αH), 5.32 (multiplet 6H) ppm. Acetone washed off 0.17 g (1.4%) (IVa) of R_f 0.06 in system 1. The oily residue was dissolved in methanol, added dropwise to acetic acid, and stored for 2 h at room temperature. (VIb) of mp 142-143° (from methanol, R_f 0.26 in system 1) was filtered off [7].

B. Without azeotropic removal of water. After the acetylation of 10 g solasodine 4.12 g (II) of mp 158-160° was obtained in a manner similar to that indicated above. On chromatographic resolution of the residue after (II) had been separated, the following were isolated: 4.98 g (II) mp 158-160° (from acetone), 0.95 g (7.9%) (VII) mp 175-176° (from ethyl alcohol), and 0.87 g (7.2%) (VIa) isolated as (IVb) mp 142-143° (from acetone).

C. Acetylation of (I) containing no water of crystallization. After the acetylation of 10 g solasodine analogously to that indicated above, 4,02 g (II) of mp 157-160° was obtained. On chromatographic resolution of the residue the following were isolated: 5.02 g (II) mp 158-160°, 0.92 g (7.6%) (VII) mp 174.5-175.5° (from ethyl alcohol), and 0.85 g (6.9%) (VIa) mp 141.5-145° (from acetone) [5]. NMR spectrum: δ0.75, 0.88, 0.99 (CH_3 groups), 1.93, 1.99 (singlet, CH_2CO ester and amide), 3.09 (multiplet, $CH_2C{=}O$), 4.53 (multiplet 3αH, 16αH), 5.31 (multiplet, 6H), and 5.98 (NH) ppm.

D. Acetylation of (I) with 80% acetic anhydride. From 9.97 g oily residue obtained after acetylating 8.5 g (I), 1.5 g (II) of mp 159-160° (from acetone) was isolated. The residue (8.47 g) from the acetone mother liquor was chromatographed on 90 g silica gel. Fraction I: chloroform (160 ml) washed off 1.87 g (II) of mp 155-157° (from acetone). Fraction II: chloroform (140 ml) washed off 1.95 g of a mixture of (II) and (X), R_f 0.24 and 0.13 in system 4. Fraction III: chloroform (180 ml) and a mixture of chloroform and ethyl acetate (5:1, 80 ml) washed off 1.5 g (15%) (III), R_f 0.28 in system 3. After two recrystallizations from acetone and ethyl acetate 0.56 g (III) of mp 130-132° [2] was obtained. Fraction IV: a mixture of chloroform and ethyl acetate (1:1, 100 ml) and ethyl acetate (100 ml) washed off 1.6 g (16%) (VII) of mp 175-178° (from ethyl alcohol). The oil (195 g) from fraction II was recrystallized from acetone and 0.6 g (II) of mp 158.5-162° was obtained. The oil (1.1 g) from the acetone mother liquor was chromatographed again on 40 g silica gel. The first portion of a mixture of cyclohexane and acetone (10:1, 60 ml) washed off 0.29 g (II) of mp 156-158°. A second portion (80 ml) of this mixture washed off 0.37 g mixture of (II) and (X) (R_f 0.24 and 0.13 in system 4). A third portion (40 ml) of this mixture washed off 0.15 g (1.5%) oil (X), R_f 0.13 in system 4. (X) was not isolated in crystalline form but was converted into (VII) [5]. (VII), 0.05 g, of mp 173-176° was obtained.

26-Acetylamino-Δ^5-cholestene-3β-16β-diol-22-one 3,16-Diacetate (VII) Treated under the Conditions for Transforming Solasodine O,N-Diacetate (II). Three grams (VII) was boiled in 40 ml glacial acetic acid for 30 min. The solution was cooled and to it at 20° was added a solution of 1.26 g sodium dichromate in 8 ml acetic acid in one lot. No increase in temperature was observed. After stirring for 10 min, 1.8 g sodium sulfite was added and the solution boiled for 3 h. Acetic acid (24 ml) was distilled off. Water (24 ml) was added at 25° with stirring, the oily precipitate extracted with methylene chloride, the extract washed until a neutral reaction was obtained and then evaporated to dryness. The residue (3.2 g of R_f 0.16 in system 1) was recrystallized from methanol and 2.4 g (VII) of mp 176-177° was obtained.

26-Acetylamino-Δ^5-furostene-3β,22-diol 3-Acetate (VIa). A. Under conditions for transforming solasodine O,N-diacetate (II). Three grams (VIa) was boiled for 30 min in 38 ml glacial acetic acid. Subsequently 1.32 g (63.5%) DPA of mp 170-172° (from ethylalcohol) [2] was obtained.

B. Under conditions for transforming ψ-isolated O,N- diacetate (III). To 5 g (VIa) in 63 ml glacial acetic acid at 20° was added with stirring 2.08 g sodium dichromate in 13 ml acetic acid in one lot. The temperature of the reaction mixture rose gradually to 33° in the course of 10 min. After 10 min stirring, 0.6 g sodium sulfite was added and the solution boiled for 3 h. Acetic acid (32 ml) was distilled off, then 32 ml water added with stirring at 25°, the precipitate filtered off and washed with 40 ml 50% aqueous acetic acid. (VIII), 1.74 g, of mp 191-205° and R_f 0.39 in system 4 was obtained. Two recrystallizations from ethyl alcohol gave 1.1 g (VIII) of mp 208-210° [7]. Mass spectrum (m/e, ε in %): M-AcOH (326, 0.61), M-AcOH-CH_3 (311, 0.12). NMR spectrum: δ 0.60, 0.82 (singlet 18-CH_3, 19-CH_3) 1.13 (doublet, 21-CH_3), 1.94 (singlet CH_5CO ester), 4.77 (multiplet, 3αH, 16αH), 5.20 (6H) ppm.

Acetylation of ψ-Solasodine-B 3,16-Diacetate (XI). Two grams (XI) was acetylated [2]. The oily residue (2 g) of R_f 0.13 in system 4 was chromatographed on 40 g silica gel. A mixture of methylene chloride and ethyl acetate (9:1) washed off 1.85 g oil (X). After rubbing under a mixture of cyclohexane, acetone,

and ether, 0.37 g (X) was filtered off with mp 164-166.5° (from a mixture of cyclohexane and acetone [5]. Ethyl acetate washed off 0.21 g (XI) of mp 198-199° (from acetone).

Study of the DPA Mother Liquors. A. ψ-Solasodine O,N-diacetate (III) (7 g) was oxidized [2]. DPA (3.23 g: 65%) of mp 170-171.5° (from ethyl alcohol) was isolated. The acetic acid mother liquor was extracted with methylene chloride; the extract was washed until it gave a neutral reaction and combined with the alcohol mother liquor from the recrystallization of technical DPA; the combined mother liquor was evaporated, and the oily substance (2.2 g) chromatographed on 80 g silica gel. A mixture of methylene chloride and chloroform (1:1) washed off 0.2 g DPA of mp 170-172°. A mixture of chloroform and ethyl acetate (10:1, 80 ml) washed off 0.27 g of a mixture of substances (R_f 0.77 and 0.49 in system 1). The remaining 160 ml of this mixture washed off 0.57 g oil of R_f 0.49 in system 1, which was acetylated by the usual method [10]. After acetylation 0.52 g oil was again chromatographed on 50 g silica gel. A mixture of benzene and ethyl acetate (4:1) washed off 0.03 g DPA of mp 170-172°. A mixture of benzene and ethyl acetate (2:1) washed off 0.12 g (IX) of mp 239-241° (with decomposition; from a mixture of hexane and acetone, 6:4). Found %: C 71.69; H 8.49; N 0.0. $C_{24}H_{34}O_5$. Calculated, %: C 71.70; H 8.45. IR spectrum: 3400 (OH), 1780 (CO lactone), 1750, 1735 (CO ester) cm^{-1} (suspension in Nujol), 3600, 1780, 1730 cm^{-1} (4% solution in chloroform). Mass spectrum (m/e, ε in %), M-AcOH (342, 0.96), M-AcOH-CH_3 (327, 0.16). NMR spectrum: δ 0.77, 0.97 (singlet 18-CH_3, 19-CH_3), 1.54 (singlet, 21-CH_3), 1.98 (singlet, CH_3CO ester), 4.55, 5.03 (3αH. 16αH), 5.31 (multiplet, 6H) ppm (in deuterochloroform).

B. In a similar manner 5.4 g oil, obtained after evaporation of the alcohol mother liquors from the recrystallization of 54 g normal technical DPA, was chromatographed on 80 g silica gel. DPA (1.91 g) of mp 170-172° (from alcohol) was isolated. Fractions containing 1.17 g of R_f 0.15 in system 4 gave, after a second chromatography on 80 g silica gel, 0.85 g (IX) of mp 239-241° (from a mixture of cyclohexane and acetone, 8:2).

C. Technical DPA (10 g) mp 156-162° was recrystallized from ethyl alcohol and 7.3 g DPA of mp 170-173° was obtained. Losses on recrystallization amounted to 27%. The mother liquor was evaporated and 2.7 g oil obtained was chromatographed on 135 g aluminum oxide (activity grade II). A mixture of benzene and ethyl acetate (10:1, 60 ml) washed off 0.14 g of a mixture of DPA and (VIII) (R_f 0.70 and 0.59 in system 2). Subsequently, 80 ml of this mixture washed off 0.12 g (VIII) of mp 203-209° (from ethyl alcohol). A mixture of benzene and ethyl acetate (5:1) washed off 0.15 g substance (R_f 0.34 in system 2) from which, after recrystallization from a mixture of hexane and acetone (6:4), 0.08 g (Vb) of mp 208-209° was obtained [11].

D. The oil (2.6 g) from the alcohol mother liquors from two recrystallizations of technical DPA, which itself had been obtained by the acetylation of 10 g (I) with 80% acetic anhydride, was chromatographed on 80 g silica gel. A mixture of benzene and ethyl acetate (10:1, 100 ml) washed off 0.59 g DPA. Subsequently 150 ml of this mixture washed off 0.17 g (VIII) of R_f 0.39 in system 4 having mp 203-205° (from ethyl alcohol). Chloroform washed off 0.1 g of a mixture of (Vb) and (IX) (R_f 0.44 and 0.37 in system 3) of mp 193-196° (from a mixture of hexane and acetone, 6:4). The mp of a mixed sample of (Vb) and (IX) was 193-220°. A mixture of chloroform and ethyl acetate (1:3) washed off 0.85 g (VII) of mp 177-179° (from ethyl alcohol).

LITERATURE CITED

1. Author's Certificate No. 118,499; Byull. Izobret., No. 6, 10 (1959).
2. N. N. Suvorov and L. M. Morozovskaya, Zh. Obshch. Khim., 30, 2062 (1960).
3. L. I. Klimova, L. M. Morozovskaya, and N. N. Suvorov, Khim.-Farmats. Zh., No. 6, 24 (1968).
4. Y. Sato, N. Ikekawa, and E. Mosettig, J. Org. Chem., 25, 783 (1960).
5. Y. Sato and N. Ikekawa, J. Org. Chem., 25, 786 (1960).
6. P. Bite and P. Tuzson, Acta Chim. Acad. Sci. Hung., 17, 241 (1958).
7. Y. Sato and N. Ikekawa, J. Org. Chem., 25, 789 (1960).
8. Y. Sato, H. Latham, and E. Mosettig, J. Org. Chem., 22, 1496 (1957).
9. N. N. Suvorov, L. V. Sokolova, and L. M. Morozovskaya, Zh. Obshch. Khim., 29, 329 (1959).
10. L. I. Klimova, L. V. Sokolova, and N. N. Suvorov, Khim. Prir. Soedin., 4, No. 5, 289 (1968).
11. U. S. Patent No. 2,623,053; Chem. Abs. 47, 10016 g (1953).