[CONTRIBUTION FROM THE RESEARCH LABORATORIES OF THE GLIDDEN COMPANY, SOYA PRODUCTS DIVISION]

Sterols. XI. 17α-Hydroxy-11-desoxycorticosterone (Reichstein's Substance S)

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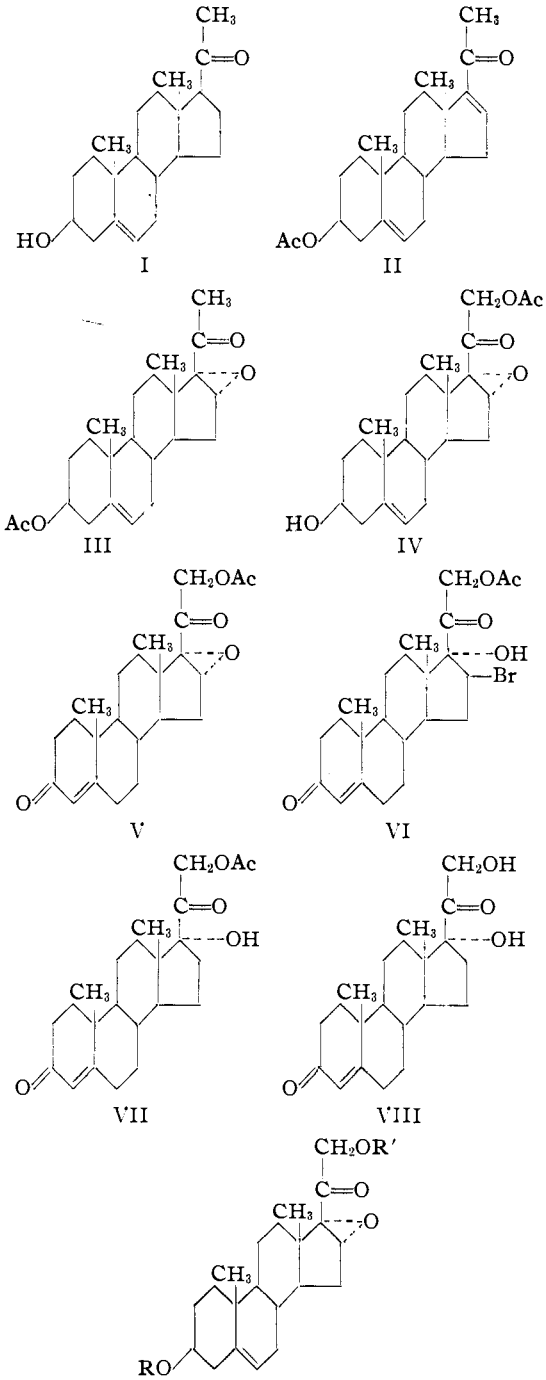
Until quite recently, 17α-hydroxy-11-desoxycorticosterone (Reichstein's Substance S (VIII)) had been the subject of comparatively little chemical or biological study since its isolation[1] and partial synthesis[2] by Reichstein and his associates. In 1946 Sarett did effect, in none too satisfactory yield, a second partial synthesis,[3] modelled after his first synthesis of Kendall's Compound E, and comprising the chromic acid oxidation of 4-pregnene-17α,20β,21-triol-3-one 21-monoacetate.[4] For large-scale preparation, however, neither of these partial syntheses was attractive, and—in the absence of any concrete and extended demand—Substance S was not available.

With the discovery of the remarkable therapeutic value of Compound E, there was an immediate expression of widespread interest in Substance S. Since, prior to the report of this discovery, we were actively engaged in a study of methods designed to make 17α-hydroxysteroids more readily available[5] for chemical and biological studies, we were constrained to devote more intensive effort to the large-scale preparation of Substance S. Starting material for our synthesis was 5-pregnene-3β-ol-20-one (I), the suitability of which resides not only in its ready availability in quantity from soya sterols but also in the fortunate presence of the 5,6-double bond.

The facile conversion of pregnenolone (I) into 5,16-pregnadiene-3β-ol-20-one acetate (II) has already been described.[6] Our more recent investigations have shown that the yields in this and similar conversions can be made even more attractive. In our first experiments leading to the synthesis of 17α-hydroxyprogesterone[7] and Substance S, the pregnadienolone acetate (II) was converted into 16,17-oxido-5-pregnene-3β-ol-20-one acetate (III) in approximately 55% yield with perbenzoic acid. Later work showed that this conversion to III could be effected in nearly quantitative yield through the use of alkaline hydrogen peroxide.

The oxidopregnenolone acetate (III) was treated first with one molar equivalent of bromine to saturate the 5,6-double bond, then with hydrogen bromide to open up the oxide, followed by a second molar equivalent of bromine for the introduction of the 21-bromo group. The resulting bromohydrin was then hydrolyzed with hydrogen bromide in methanol to yield the corresponding 3-hydroxy compound; the latter



(1) Reichstein and von Euw, *Helv. Chim. Acta*, **21**, 1197 (1938); Reichstein, *ibid.*, **21**, 1490 (1938).
(2) Reichstein and von Euw, *ibid.*, **23**, 1258 (1940).
(3) Sarett, *J. Biol. Chem.*, **162**, 627 (1946).
(4) *Cf.* Ruzicka and Müller, *Helv. Chim. Acta*, **22**, 755 (1939).
(5) Julian, Meyer and Ryden, THIS JOURNAL, **71**, 756 (1949).
(6) Julian and Karpel, *ibid.*, **72**, 362 (1950).
(7) (a) Julian, Meyer, Karpel and Ryden, *ibid.*, **71**, 3574 (1949); (b) Julian, Meyer and Ryden, *ibid.*, **72**, 367 (1950).

was then treated with sodium iodide in benzene–ethanol resulting in regeneration of the 5,6-double bond and replacement of bromine at C_{21} by iodine, and the crude 21-iodo product converted by the action of potassium acetate in acetone, to 16,17-oxido-5-pregnene-3β,21-diol-20-one 21-acetate (IV). It proved to be unnecessary to isolate and purify the intermediates in the successive stages down to IV. Oppenauer oxidation of IV yielded 16,17-oxido-4-pregnene-21-ol-3,20-dione acetate (V), which, upon treatment with hydrogen bromide in acetic acid, gave the bromohydrin (VI). Dehalogenation of VI with Raney nickel afforded 17α-hydroxy-11-desoxycorticosterone acetate (VII). Hydrolysis of VII with potassium bicarbonate in aqueous methanol gave the desired Substance S.

For purposes of characterization and extension of this method of synthesis, the derivatives IX, X and XI were prepared. The above outlined synthesis of Substance S, applicable to comparatively large quantities, is characterized by many elements of simplicity and convenience, among which mention should be made of the economical introduction of oxygen at C_{17} with retention of the 5,6-double bond intact. The elegant synthesis of Gallagher and his associates[8] which was recorded in a communication appearing slightly prior to our own first record,[7] employs pregnane-3α-ol-20-one as starting material, thus involving the unwelcome task of introduction of the double bond into Ring A.

Experimental[9]

16,17-Oxido-5-pregnene-3β-ol-20-one Acetate (III).—Three grams of 5,16-pregnadiene-3β-ol-20-one acetate (II) was dissolved in 200 ml. of methanol. This solution was treated, after chilling to 15°, with 6 ml. of 4 *N* sodium hydroxide solution and then immediately with 12 ml. of 30% hydrogen peroxide solution. The mixture was then stored in the refrigerator at 5° for twenty-three hours. At this time, examination of aliquot samples indicated the absence of an α,β-unsaturated ketone as evidenced by absorption in the ultraviolet and the consumption of approximately one molar equivalent of hydrogen peroxide determined by permanganate titration. The methanol solution was poured into 800 ml. of water and the resulting white solid separated. The solid, which was washed well with water and dried, weighed 2.63 g. (95%) and melted at 180–186°. Recrystallization of a sample from methanol gave colorless needles melting at 187–190° which showed no depression in melting point when admixed with an authentic sample of 16,17-oxido-5-pregnene-3β-ol-20-one.[7b] Acetylation of the 3β-hydroxy compound with acetic anhydride in dry pyridine gave the known acetate (III).[7b]

16,17-Oxido-5-pregnene-3β,21-diol-20-one 21-Acetate (IV).—A solution of 10.0 g. of 16,17-oxido-5-pregnene-3β-ol-20-one acetate[7] in 200 ml. of acetic acid–carbon tetrachloride (1:1), chilled to 18°, was treated with a solution of 4.3 g. of bromine in 30 ml. of carbon tetrachloride. Upon complete decolorization, there was added 15 ml. of a 32% solution of hydrogen bromide in acetic acid. After the solution had stood for ten minutes at room temperature, a second molar equivalent of bromine, 4.3 g. in 30 ml. of carbon tetrachloride, was added portionwise at room temperature with stirring during a forty-minute period. The reaction mixture was allowed to stand for an additional fifteen minutes and then evaporated *in vacuo* with a minimum of heating to remove the carbon tetrachloride. The remaining suspension was poured into water, filtered and the separated solid washed with water and dried at 50°. The solid, 17.9 g., was dissolved in 75 ml. of benzene and 175 ml. of methanol and after the addition of 5.2 g. of hydrogen bromide in 15 ml. of methanol, the solution was allowed to stand at room temperature for ten hours. The reaction mixture was diluted with water and extracted with ether. The ethereal extract was then washed with water, dried, concentrated to 20 ml. and diluted with 155 ml. of benzene. After the addition of a solution of 36.5 g. of sodium iodide in 175 ml. of absolute ethanol, the mixture was allowed to stand at room temperature for twenty-two hours. It was then diluted well with water and extracted with ether. The extract was washed with 3% sodium thiosulfate solution, then with water and dried. The cream-colored residue (14.5 g.) remaining after removal of solvent *in vacuo* was dissolved in 300 ml. of acetone containing 42.0 g. of freshly-fused potassium acetate. The mixture was refluxed for four and one-half hours, then concentrated to a small volume, diluted with water and extracted with ether. The water-washed and dried ethereal solution gave upon concentration to a small volume, 5.5 g. (53%) of 16,17-oxido-5-pregnene-3β,21-diol-20-one 21-acetate melting at 180–188°. A slurry of the acetate in ether yielded 5.0 g. of material melting at 188–190°. Further recrystallization from acetone gave colorless needles melting at 190–192°: $[\alpha]^{24}D$ + 14.9° (8.7 mg. made up to 2 ml. with chloroform, αD + 0.065°, *l*, 1 dm.).

Anal. Calcd. for $C_{23}H_{32}O_5$: C, 71.11; H, 8.30. Found: C, 70.86; H, 8.33.

Several standardized runs of the above compound were made employing in each, 300 g. of the starting oxide. The results were similar to that recorded above.

16,17-Oxido-4-pregnene-21-ol-3,20-dione Acetate (V).—A solution of 6.5 g. of aluminum isopropoxide in 60 ml. of toluene was added dropwise during a period of five minutes to a solution of 15.0 g. of 16,17-oxido-5-pregnene-3β,21-diol-20-one 21-acetate in 600 ml. of toluene and 130 ml. of cyclohexanone. The mixture was refluxed for an additional one-half hour and after the addition of 3 ml. of acetic acid in 20 ml. of toluene, it was steam distilled. The crystalline residue was dissolved in ether and washed with 1% sulfuric acid, 1% sodium hydroxide solution and then with water to neutrality. Addition of petroleum ether (b. p. 35–60°) to the dried and concentrated ethereal solution (60 ml.) gave 9.0 g. of 16,17-oxido-4-pregnene-21-ol-3,20-dione acetate in clusters of needles melting at 168–170°. From the mother-liquor there was secured an additional 1.5 g. (total yield 70%) of material melting at 165–168°. Several recrystallizations from ether–petroleum ether (b. p. 35–60°) gave prisms melting at 170–172°; $[\alpha]^{25}D$ + 166.8° (7.1 mg. made up to 2 ml. with chloroform, αD + 0.592°, *l*, 1 dm.).

Anal. Calcd. for $C_{23}H_{30}O_5$: C, 71.48; H, 7.82. Found: C, 71.82; H, 8.23.

17α-Hydroxy-11-desoxycorticosterone Acetate (VII).—A solution of 20 ml. of 32% hydrogen bromide in acetic acid was added to a chilled solution (15–18°) of 20.0 g. of 16,17-oxido-4-pregnene-21-ol-3,20-dione acetate in 100 ml. of acetic acid. The solution, red-brown in color, was held for fifteen minutes at room temperature and then cooled to 18° for an additional fifteen minutes. The crystalline bromohydrin (23.5 g., 97%) which separated within two to three minutes was filtered, washed with alcohol-free ether and dried *in vacuo* for five-ten minutes at 45°. In one experiment, a sample of the bromohydrin (VI) when recrystallized from benzene–ether formed needles melting at 177–178° dec.

Anal. Calcd. for $C_{23}H_{31}O_5Br$: C, 59.10; H, 6.73. Found: C, 59.26; H, 7.05.

A solution of 23.5 g. of the crude bromohydrin in 750 ml.

(8) Koechlin, Garmaise, Kritchevsky and Gallagher, THIS JOURNAL, **71**, 3262 (1949).

(9) Carbon and hydrogen analyses by Micro-Tech Laboratory, Skokie, Illinois.

of 95% ethanol was stirred and refluxed with 98 g. of Raney nickel[10] for five hours and then filtered while hot through a bed of filter-aid. The pale yellow filtrate was concentrated *in vacuo* to a slurry of needle-like crystals and chilled. The white solid was separated, washed with 100 ml. of cold acetone and dried. There was obtained 13.5 g. (67%) of 17α-hydroxy-11-desoxycorticosterone acetate melting at 228–232°. Recrystallization from acetone gave colorless needles melting at 235–238° (sinters at 230°). This material showed no depression in melting point when admixed with a sample of the acetate derived from a natural source[11]; $[\alpha]^{24}D$ + 114° (11.2 mg. made up to 5 ml. with acetone, αD + 0.255°, *l*, 1 dm.).

Anal. Calcd. for $C_{23}H_{32}O_5$: C, 71.11; H, 8.30. Found: C, 70.94; H, 8.28.

This material when dissolved in concentrated sulfuric acid produced the typical scarlet color.

17α-Hydroxy-11-desoxycorticosterone (VIII).—A solution of 500 mg. of 17α-hydroxy-11-desoxycorticosterone acetate in 75 ml. methanol containing 500 mg. of potassium bicarbonate in 10 ml. of water was allowed to stand at room temperature for twenty-four hours. It was then diluted with saline solution and extracted with freshly-distilled ether. The ether solution was washed with water to neutrality, dried and concentrated to 10 ml. After chilling, the separated solid was filtered and washed with cold ether. There resulted 390 mg. of fine, glistening plates melting at 200–208° (87%). Recrystallization of this material from acetone raised the melting point to 207–208°.[12]

16,17-Oxido-5-pregnene-3β,21-diol-20-one 3,21-Diacetate (IX).—To a solution of 10.0 g. of 16,17-oxido-5-pregnene-3β-ol-20-one acetate in 100 ml. of acetic acid chilled in an ice-bath, a solution of 4.3 g. of bromine in 11.5 ml. of acetic acid was added with swirling. Upon disappearance of the bromine color, the solution was treated with 15 ml. of 32% hydrogen bromide in acetic acid. The solution became red-brown in color and a white solid separated. After one-half hour at room temperature, 150 ml. of carbon tetrachloride was added and the mixture was warmed gently to effect complete solution. A second molar equivalent of bromine (4.3 g. in 11.5 ml. of acetic acid) was added. After forty-five minutes, the yellow solution was concentrated *in vacuo* at a temperature not exceeding 45° until a heavy crystalline slurry remained. The mixture was diluted with water, filtered and the cake was washed well with water and dried. The crude tetrabromo compound (turns brown at 130° and decomposes rapidly at 145–147°) was dissolved in 250 ml. of benzene and then treated with a solution of 40.0 g. of sodium iodide in 250 ml. of ethanol. After standing at room temperature for twenty-one hours, the mixture was poured into 2 l. of water and extracted with ether. The ethereal layer was washed successively with dilute sodium thiosulfate solution and water, dried and concentrated *in vacuo*. The tan, solid residue was dissolved in 300 ml. of acetone and after the addition of 30.0 g. of freshly-fused potassium acetate, the mixture was refluxed for three hours. It was then diluted with water and extracted with ether. The ether extract was washed several times with water, dried and concentrated. Crystallization of the residue from methanol afforded 7.7 g. (67%) of white needles melting at 170–175°. Several recrystallizations from the same solvent raised the melting point to 175–177°; $[\alpha]^{28}D$ + 10.2° (23.2 mg. made up to 2.0 ml. with chloroform, αD + 0.118°, *l*, 1-dm.).

Anal. Calcd. for $C_{25}H_{34}O_6$: C, 69.74; H, 7.96. Found: C, 69.83; H, 8.01.

16,17-Oxido-5-pregnene-3β,21-diol-20-one 3-Acetate 21-Benzoate (X).—In much the same fashion as described above, but employing one-tenth of the stated amounts of reagents and carrying out the last step with 3.0 g. of potassium benzoate, 1.0 g. of 16,17-oxido-5-pregnene-3β-ol-20-one acetate was converted to 710 mg. (54%) of the 21-benzoxy derivative (crystallized from chloroform–methanol) melting at 188–190°. Several recrystallizations from the same solvent pair gave long, silky needles melting at 190–191°.

Anal. Calcd. for $C_{30}H_{36}O_6$: C, 73.14; H, 7.36. Found: C, 72.94; H, 7.57.

16,17-Oxido-5-pregnene-3β,21-diol-20-one 3-Formate 21-Acetate (XI).—A solution of 500 mg. of 16,17-oxido-5-pregnene-3β-ol-20-one in 5 ml. of chloroform was treated with a solution of 245 mg. of bromine in 2.45 ml. of chloroform. After the bromine color had disappeared, 2 ml. of 20% hydrogen bromide in formic acid was added. The reaction mixture which separated into two phases was swirled occasionally during one-half hour. An additional 245 mg. of bromine in 2.45 ml. of chloroform was added and the mixture was allowed to stand, with occasional swirling, at room temperature until the bromine color had disappeared (about two hours). The reaction mixture was diluted with ether and water. The ethereal layer was washed with water, dilute sodium bicarbonate solution and then water until neutral. Concentration of the dried solution *in vacuo* gave a resinous mass. This amorphous tetrabromide was then treated as described above with proportional amounts of sodium iodide in ethanol–benzene followed by potassium acetate in acetone. Crystallization of the crude reaction product from ether–petroleum ether (b. p. 35–60°) gave 245 mg. (39%) of material melting at 168–174°. Several recrystallizations from the same solvent pair produced fine, white needles melting at 180–182°.

Anal. Calcd. for $C_{24}H_{32}O_6$: C, 69.21; H, 7.75. Found: C, 69.00; H, 7.71.

Summary

A new partial synthesis of 17α-hydroxy-11-desoxycorticosterone (Reichstein's Compound S) is described.

CHICAGO, ILLINOIS RECEIVED JUNE 5, 1950

(10) Prepared from Raney alloy as described by Adkins, "Reactions of Hydrogen," University of Wisconsin Press, Madison, Wisconsin, 1937.

(11) We are indebted to Dr. Marvin H. Kuizenga of The Upjohn Laboratories for a sample of this material.

(12) Reichstein, *Helv. Chim. Acta*, **21**, 1490 (1938).