

Steroidal Sapogenins. LIII. Permanganate-Periodate Oxidation of Pseudosapogenins²

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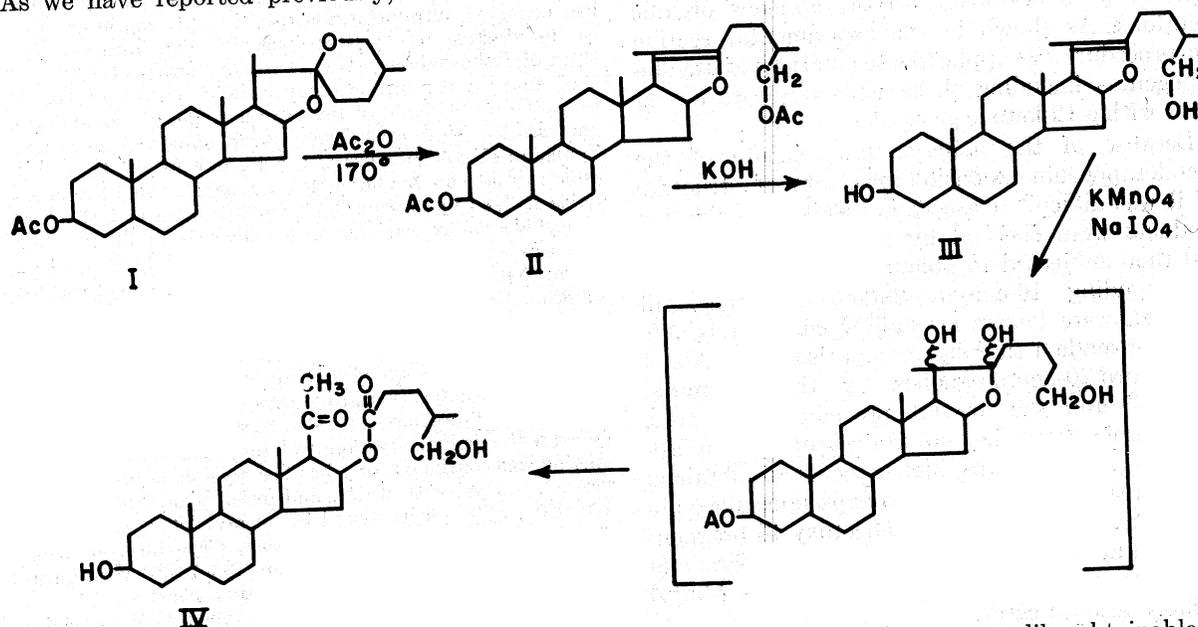
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Pseudosapogenins and certain other steroid olefins with especially reactive double bonds can be smoothly oxidized with a permanganate-periodate reagent.

Modern steroid hormone technology is based in large part on 16-dehydro-20-oxo-pregnenes obtained by oxidative degradation of the steroidal sapogenin side chain. The classical three-step process was first developed by R. E. Marker and his associates³ consisting of (1) conversion of sapogenins to pseudosapogenins, (2) oxidation of pseudosapogenins to give 16 β -acyl esters of 20-oxo-pregnanes, and (3) alkaline cleavage of the 16 β -ester followed by dehydration to give 16-dehydro-20-oxo-pregnanes. The over-all process has been studied in a number of laboratories^{4a-h} leading to a degree of improvement in the classical procedure. However, in all of the cited references the oxidation agent has been chromic acid in acetic acid. As we have reported previously,^{4c} the 16 β -esters

formed during oxidation of pseudosapogenins invariably undergo partial hydrolysis under the acidic reaction conditions giving 16-dehydro-20-oxo-pregnenes which may then further react with excess oxidant. In order to avoid this undesirable hydrolysis we have studied the oxidation of pseudosapogenins under neutral or slightly alkaline conditions. A procedure developed by Lemieux and von Rudloff⁵ involving permanganate-periodate oxidation of olefins soluble in slightly alkaline aqueous media seemed to offer promising possibilities. With some slight modifications the procedure was readily adapted to the oxidation of water insoluble pseudosapogenins by the route shown in Chart I.

The details of the procedure were developed



(1) Eastern Utilization Research and Development Division, Agricultural Research Service, U. S. Department of Agriculture.

(2) Previous paper in this series, "Steroidal Sapogenins. LII. Structure and Properties of the Acetyl Hypobromite Adduct from a Δ^{16} -Pregnen-20-one," S. G. Levine and M. E. Wall, *J. Am. Chem. Soc.*, in press.

(3) R. E. Marker *et al.*, *J. Am. Chem. Soc.*, **62**, 3350 (1940); *J. Am. Chem. Soc.*, **64**, 468 (1942); *J. Am. Chem. Soc.*, **69**, 2167 (1947).

(4) (a) D. H. Gould, H. Staeudle, and E. B. Hershberg, *J. Am. Chem. Soc.*, **74**, 3685 (1952); (b) W. G. Dauben and G. J. Fonken, *J. Am. Chem. Soc.*, **76**, 4618 (1954); (c)

with pseudosarsasapogenin, readily obtainable in pure, crystalline condition.⁶ The conversion of

M. E. Wall, H. E. Kenney, and E. S. Rothman, *J. Am. Chem. Soc.*, **77**, 5665 (1955); (d) M. E. Wall and S. Serota, *J. Am. Chem. Soc.*, **79**, 6481 (1957); (e) A. F. B. Cameron, R. M. Evans, J. C. Hamlet, J. S. Hunt, P. G. Jones, and A. G. Long, *J. Chem. Soc.*, 2807 (1955).

(5) R. U. Lemieux and E. von Rudloff, *Can. J. Chem.*, **33**, 1710 (1955).

(6) M. E. Wall, S. Serota, and C. R. Eddy, *J. Am. Chem. Soc.*, **77**, 1230 (1955).

pseudosapogenins III to the corresponding 16 β -ester, IV, was conveniently followed by noting the decrease or disappearance of the ultraviolet absorption band at 215 $m\mu$ found in pseudosapogenins,^{3e} and by observing the concomitant appearance of strong infrared absorption bands at 1725 $cm.^{-1}$ and 1707 $cm.^{-1}$ due to 16 β -ester and 20-ketone groups, respectively, found in IV. After some experimentation the correct conditions for conducting the oxidation of the water-insoluble pseudosapogenins were developed. It was found that the steroid, in the free hydroxyl form, must be dissolved in a water-miscible, inert solvent such as dioxane or tertiary butylalcohol. The steroid solution was then added to the aqueous solution of inorganic reagents and the mixture strongly agitated or shaken. The general reaction conditions of Lemieux and von Rudloff,⁵ which utilize for 1 Mm. of olefin an oxidizing mixture of 3 Mm. of potassium carbonate, 8 Mm. of sodium metaperiodate, and 0.34 Mm. potassium permanganate at room temperature, were satisfactory under our experimental conditions and gave rapid oxidation of the C₂₀, C₂₂ double bond. Due possibly to their lower solubility in the aqueous dioxane or tertiary butyl alcohol solutions, pseudosapogenin diacetates were poorly oxidized under similar conditions. The yield was not improved by adding benzene as a co-solvent to the aqueous organic solutions. As shown in the experimental section the procedure was applicable to a variety of pseudosapogenins including those with unsaturation at C₅ or with a 12-ketone group.

Because of the noncrystalline nature of the pseudosapogenin oxidation products of structure IV it was difficult to ascertain yields. All oxidation products were checked by infrared spectroscopy and then subjected to alkaline hydrolysis^{4c} to the corresponding 16-dehydro-20-keto-pregnenes, all of which were known crystalline compounds. We have not made a thorough comparison of yields of 16-dehydro-20-ketopregnenes by the chromium trioxide^{4c} and the present procedure. However, with simple cases, i.e. no Δ^5 -unsaturation or absence of 12-keto groups, higher yields are obtained by the present method. Thus from pseudosmilagenin we obtained a 40% yield of 3 β -acetoxy-16-pregnen-20-one and 70% by the permanganateperiodate procedure, using potassium hydroxide in *t*-butyl alcohol^{4c} in both cases.

Because of the mild, smooth oxidation action of the permanganateperiodate reagent we tested its effect on several types of steroidal olefins available to us. Oxidation of 12-methylene tigogenin⁷ proceeded smoothly to give hecogenin in excellent yield. Similar treatment of 3 β -hydroxy-16-pregnen-20-one converted this steroid completely to acidic products which were not further characterized. An enol-acetate, 3 β ,20-diacetoxy-17-pregnene, and stigmasterol, a steroid with both a tri- and a di-

substituted olefinic linkage, were recovered unchanged. From our brief experiences with a limited variety of steroid olefins we conclude that only highly reactive steroid double bonds are attacked by the permanganate-periodate reagent. This is not a general observation because as previously shown by Lemieux and von Rudloff the disubstituted olefinic bond in oleic acid is easily oxidized. Whether this is due to the greater solubility of sodium oleate in the aqueous medium or to steric factors which may be encountered with steroids cannot be decided by evidence on hand.

EXPERIMENTAL

All pseudosapogenins were prepared in a manner described previously.^{4d} The following oxidation procedure is typical. A solution of 1.2 g. of potassium permanganate (3.2 Mm.), 10.0 g. of anhydrous potassium carbonate (71.4 Mm.), and 41.0 g. of sodium metaperiodate (190.4 Mm.) in 1.0 l. of water was mixed with a solution of 10.0 g. (23.8 Mm.) of pseudosarsasapogenin in 500 ml. of purified dioxane and 500 ml. of benzene. The mixture was vigorously shaken for 1 hr. Probe tests on smaller quantities indicated the reaction was substantially complete in 5 min. The benzene layer was separated from the aqueous fraction, the latter extracted several times with benzene, and all the benzene extracted united and washed with distilled water. The benzene was concentrated to dryness *in vacuo* leaving a pale yellow glass which was undoubtedly 3 β -hydroxy-16 β -(γ -methyl- β -hydroxy)-valeroxy-pregnan-20-one. The compound had negligible ultraviolet absorption at 215 $m\mu$ in contrast to the starting pseudosarsasapogenin. The infrared spectrum of I showed two strong carbonyl bands at 1720 and 1707 $cm.^{-1}$ and a strong band at 1250 $cm.^{-1}$ attributable to the C—O—C bond of the 16 β -ester. Because of the well known difficulty in crystallizing compounds with structures similar to IV and because of the general instability of 16 β -ester steroids no further attempts were made to characterize it. The viscous product was taken up in 200 ml. of *t*-butyl alcohol by warming on the steam bath. To the solution was added 5.0 g. of potassium hydroxide in 4 ml. of water. Nitrogen was passed through the *t*-butyl alcohol solution. The flask was stoppered and vigorously stirred with a magnetic stirrer for 3 hr. The initial temperature was 40° which fell to 30° at the expiration of 3 hr. To the hydrolyzed solution was added 400 ml. of water with vigorous stirring. The aqueous solution was seeded with authentic 3 β -hydroxy-16-pregnen-20-one and another 400 ml. of water added with stirring. Crystalline plates began to form and the stirring was continued for 1 hr. The crystalline product was filtered, washed with water, and dried, yield 6.91 g., m.p. 170–180°, 85% purity based on ultraviolet absorption at 239 $m\mu$, equivalent to 77% of theory. Crystallization from methanol gave 5.0 g. of plates, m.p. 185–187°^{4e} with infrared spectrum identical to an authentic specimen.

In exactly the same manner described above pseudodiosgenin, pseudotigogenin, and pseudohecogenin were oxidized to their corresponding 16 β -acyl esters and hydrolytically cleaved to give respectively 3 β -hydroxy-5,16-pregnadiene-20-one, 3 β -hydroxy-5 α -pregn-16-en-20-one, and 3 β -hydroxy-5 α -pregn-16-en-12,20-dione, all characterized by melting point and identity with known samples.^{4e} The yields were not so high as in the pseudosarsasapogenin oxidation ranging from 30–50% of theory by ultraviolet absorption analysis. It was found that *t*-butyl alcohol was as effective a solvent as dioxane for this purpose and since it was much easier to purify, the former solvent was adopted for all subsequent work. It was also found that the use of benzene during oxidation was unnecessary. However no reaction occurred if an aqueous solution of the inorganic oxidant was

shaken with a benzene solution of steroid in the absence of dioxane or *t*-butyl alcohol.

Oxidation of 12-methylene-tigogenin to hecogenin. 0.4 g. of 12-methylene tigogenin⁷ in 100 ml. of *t*-butyl alcohol was oxidized with a solution of 1.7 g. of sodium metaperiodate, 0.42 g. of potassium carbonate, and 0.05 g. of potassium permanganate in 100 ml. of water, shaking the mixture in a 500-ml. bottle. Infrared analysis for carbonyl indicated maximum formation in 5 hr. The solution was extracted with benzene, yielding after the usual work-up 0.3 g. of hecogenin, m.p. 250–253°, infrared spectrum identical with that of an authentic specimen.

Oxidation of 3 β -acetoxy-16-pregnen-20-one. A solution of

(7) F. Sondheimer and R. Mechoulam, *J. Am. Chem. Soc.*, 79, 5029 (1957).

0.42 g. of 3 β -acetoxy-16-pregnen-20-one in 100 ml. of *t*-butyl alcohol was shaken overnight with the aqueous oxidation solution used above. The aqueous mixture was further diluted with water and extracted with ether. The ether solution contained a negligible weight of steroid. The aqueous layer was acidified with hydrochloric acid and the resultant precipitate extracted with ether to yield 0.4 g. of amorphous glass. The infrared absorption spectrum showed, as might be expected, a strong carboxyl carbonyl band at 1700 cm.⁻¹.

Under the above reaction conditions stigmaterol and 3 β ,20-diacetoxy-17-pregnen-20-one were recovered unchanged.

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