one hour a solution of 1.2 g. of chromium trioxide in 25 cc. of 90% acetic acid. The solution was stirred for several hours at 20° and allowed to stand overnight at 25°. Methyl alcohol (20 cc.) was added, the solution was evaporated *in vacuo* to dryness and the residue was dissolved in 250 cc. of water and 250 cc. of ether. The ether solution was washed thoroughly with water and then extracted with sodium carbonate solution and the sodium carbonate solution, after extraction with ether, was acidified to liberate an acid which was filtered off and crystallized from benzene–pentane and also from dilute acetone: m. p. 250°; yield 0.3 g. It gave no depression with the acid prepared by lead tetraacetate–hydrogen peroxide oxidation of this 4-hydroxycholestanol.[2]

Conversion to the dimethyl ester with diazomethane and crystallization from methyl alcohol gave needles, m. p. 123–124°.

Anal. Calcd. for $C_{27}H_{46}O_4$: C, 74.6; H, 10.7. Found: C, 74.5; H, 10.7.

Summary

The oxidation of sitosterol by selenium oxide gave 4-hydroxy- and 6-hydroxysitosterol. Hydrogenation of the 4-hydroxy compound gave 4-hydroxysitostanol. 4-Hydroxycholestanol upon oxidation with chromic acid gave the same dicarboxylic acid as upon treatment with lead tetraacetate. An analogous dicarboxylic acid was obtained upon the oxidation of 4-hydroxysitostanol by chromic acid.

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[CONTRIBUTION FROM THE SCHOOL OF CHEMISTRY AND PHYSICS OF THE PENNSYLVANIA STATE COLLEGE]

Sterols. XXXII. Oxidation of Stigmasterol by Selenium Oxide

BY RUSSELL E. MARKER AND EWALD ROHRMANN

Rosenheim and Starling[1] studied the oxidation of cholesterol by selenium dioxide in acetic acid solution and reported the formation of two stereoisomers, namely, *cis*-4-hydroxycholesterol and *trans*-4-hydroxycholesterol. Butenandt and Hausmann,[2] however, showed that the assumed *trans*-4-hydroxycholesterol actually is 6-hydroxycholesterol.

We have now extended this oxidation to stigmasterol and stigmasteryl acetate and find that the reaction is similar to that with cholesterol. In analogy to the reactions in the cholesterol series, the diacetate of 4-hydroxystigmasterol is less soluble than the diacetate of the 6-hydroxy compound while with the free diols the solubility relationship is reversed. These factors make possible an easy separation of the two diols. The oxidations were carried out in acetic acid in the presence of benzene at about 90°. Contrary to the results reported by Rosenheim and Starling[1] for cholesterol, we find that the oxidation of the acetate of stigmasterol readily yields the monoacetate of 4-hydroxystigmasterol. The main reactions involved in this study are illustrated in the chart.

Hydrogenation of the diacetate of 4-hydroxystigmasterol with platinum oxide catalyst yielded the diacetate of 4-hydroxystigmastanol which proved to be identical with the diacetate of 4-hydroxysitostanol.[3] Reduction of the diacetate of 4-hydroxystigmastanol by the method of Clemmensen gave a hydrocarbon which was identical with sitostane prepared by the action of sodium on sitostyl chloride. Hydrolysis of the diacetate of 4-hydroxystigmastanol yielded 4-hydroxystigmastanol which was identical with 4-hydroxysitostanol. These facts give further evidence that stigmasterol differs from sitosterol only in the presence of a double bond in the side chain.

The microanalyses herein reported have been performed by Dr. George H. Fleming.

Experimental Part

Diacetate of 4-Hydroxystigmasterol.—To a solution of 10 g. of stigmasteryl acetate in 50 cc. of benzene was added a hot solution of 4 g. of selenium dioxide in 100 cc. of 98% acetic acid. The solution was refluxed for one hour, then 10 g. of sodium acetate was added and refluxing continued for ten minutes to coagulate the selenium. The mixture then was poured into 200 cc. of water and the benzene layer was separated. After removal of the solvent the residue was dissolved in 30 cc. of acetic anhydride and heated under a reflux condenser during one-half hour. The acetyl derivative which separated on cooling was filtered off, dissolved in ether (100 cc.) and treated with decolorizing charcoal (Norite). After filtration and partial evaporation of the ether the crystalline product was obtained by the addition of 200 cc. of cold

(1) Rosenheim and Starling, *J. Chem. Soc.*, 377 (1937).
(2) Butenandt and Hausmann, *Ber.*, **70**, 1154 (1937).
(3) Marker, Kamm and Wittle, THIS JOURNAL, **60**, 1071 (1938).

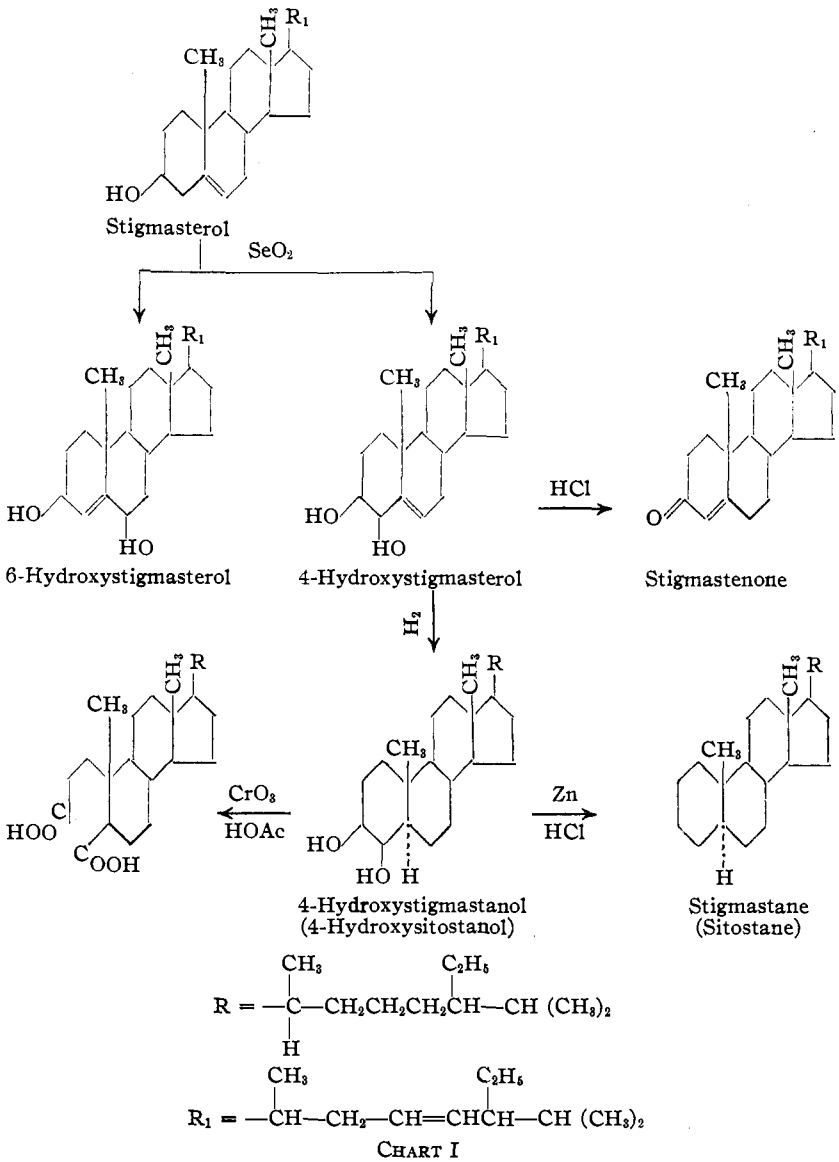


CHART I

methanol. Repeated crystallization from acetone and ethyl acetate yielded a product melting at 200–201°.

Anal. Calcd. for $C_{33}H_{52}O_4$: C, 77.3; H, 10.2. Found: C, 77.0; H, 10.3.

3-Acetoxy-4-hydroxystigmasterol.—This product was prepared by the method given above for the diacetate except that the acetic anhydride treatment was omitted. The benzene layer was concentrated to crystallization and filtered. The dark colored crystals were dissolved in a mixture of equal parts of ether and acetone, the solution being treated with Norite, filtered, and concentrated to crystallization. Repeated crystallization from ether–acetone and acetone–ethyl acetate mixtures yielded white plates melting at 193–195°.

Anal. Calcd. for $C_{31}H_{50}O_3$: C, 79.1; H, 10.6. Found: C, 79.6; H, 10.8.

When heated with acetic anhydride this 3-acetoxy derivative was converted into the diacetate melting at 198–200°.

4-Hydroxystigmasterol.—A solution of 2 g. of the diacetate of 4-hydroxystigmasterol in 200 cc. of alcohol with an excess of potassium hydroxide was refluxed for thirty minutes. Water was added and the product was extracted with ether. After removal of the solvent and crystallization from acetone the product melted at 188°.

Anal. Calcd. for $C_{29}H_{48}O_2$: C, 81.2; H, 11.3. Found: C, 80.9; H, 11.4.

4-Hydroxystigmastanol Diacetate.—A solution of 2 g. of the diacetate of 4-hydroxystigmasterol in 200 cc. of acetic acid containing 500 mg. of platinum oxide was shaken with hydrogen at 45 pounds (3 atm.) pressure for one hour. The catalyst was filtered off, the acetic acid removed by vacuum distillation and the product crystallized from ether–ethanol and acetone. It melted at 153° and when mixed with the diacetate of 4-hydroxysitostanol gave no depression in melting point.

Anal. Calcd. for $C_{33}H_{56}O_4$: C, 76.7; H, 10.9. Found: C, 76.9; H, 10.9.

4-Hydroxystigmastanol.—A solution of 500 mg. of the diacetate of 4-hydroxystigmastanol was hydrolyzed with alcoholic potassium hydroxide solution. The product was crystallized from ether (in which it is sparingly soluble), ethanol, and acetone. It melted at 203° and gave no depression in melting point when mixed with 4-hydroxysitostanol.

Stigmastenone from 4-Hydroxystigmasterol.—A solution of 1 g. of 4-hydroxystigmasterol in 100 cc. of ethanol containing 5 cc. of concd. hydrochloric acid was refluxed for ten minutes. After removal of the alcohol the product which remained was crystallized from ether–methanol, acetone, and ethyl acetate. It then melted at 94° and gave no depression in melting point when mixed with stigmastenone.

Stigmastane from the Diacetate of 4-Hydroxystigmastanol.—A solution of 1 g. of the diacetate of 4-hydroxystigmastanol in a mixture of acetic acid and hydrochloric acid containing 15 g. of amalgamated zinc was refluxed for three hours. The product was extracted with ether and then sublimed in high vacuum at 100°. After crystallization from alcohol–ether it melted at 84° and gave no depression in melting point when mixed with sitostane pre-

pared by the action of sodium and amyl alcohol on sitostyl chloride.

Anal. Calcd. for $C_{29}H_{52}$: C, 86.9; H, 13.1. Found: C, 87.2; H, 13.0.

6-Hydroxystigmasterol.—The acetic anhydride filtrate from the preparation of the diacetate of 4-hydroxystigmasterol was evaporated to dryness *in vacuo*. The residue was dissolved in 200 cc. of ethyl alcohol, refluxed for one hour with 6 g. of potassium hydroxide, and the solution then acidified with acetic acid. On dilution with water a product separated and was filtered off, dissolved in ethyl alcohol and treated with Norite. Evaporation and cooling of the alcohol gave crystals which on recrystallization from ethyl acetate gave 6-hydroxystigmasterol in long needles melting at 237°.

Anal. Calcd. for $C_{29}H_{48}O_2$: C, 81.2; H, 11.3. Found: C, 80.6; H, 11.2.

Summary

4-Hydroxy- and 6-hydroxystigmasterol have been prepared by the oxidation of stigmasterol with selenium dioxide in acetic acid–benzene solution. A similar oxidation of stigmasteryl acetate yielded 3-acetoxy-4-hydroxystigmasterol.

Hydrogenation of the diacetate of 4-hydroxystigmasterol yielded the diacetate of 4-hydroxystigmastanol which is identical with the diacetate of 4-hydroxysitostanol. Clemmensen reduction of the diacetate of 4-hydroxystigmastanol yields stigmastane which is identical with sitostane.

4-Hydroxystigmasterol when heated with alcoholic hydrochloric acid undergoes dehydration and rearrangement to form stigmastenone.

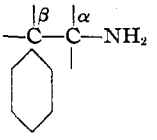
STATE COLLEGE, PENNA. RECEIVED FEBRUARY 25, 1938

[CONTRIBUTION FROM THE RESEARCH LABORATORIES OF THE UPJOHN COMPANY]

Physiologically Active Phenethylamines. II. Hydroxy- and Methoxy-β-methyl-β-phenethylamines (β-Phenyl-*n*-propylamines)

BY E. H. WOODRUFF AND EARL PIERSON[1,2]

The number of individual amines related structurally and pharmacologically to epinephrine and ephedrine that have been synthesized is large, numbering over four hundred at the present time. All possess the basic skeleton



which Barger and Dale[3] found to be necessary for sympathomimetic activity. The simplest compound possessing activity is that in which the valences of the carbon atoms in the side chain are satisfied with hydrogen. The substitution of alkyl groups in place of one of the hydrogen atoms on the α-carbon, in particular the methyl group, has been investigated extensively and a wide variety of compounds possessing this skeleton has been synthesized.[4]

The effect of alkyl groups on the β-carbon has received on the contrary no such strenuous attention. The only mention of this class of amines in the literature is confined to the simple β-methyl-β-phenethylamine.[5–7] Hartung and Munch report briefly on its properties in a study of the isomeric phenylpropylamines. In this work they state that it is orally active and possesses sympathomimetic activity. It is with the intention of more completely investigating the pharmacology of this hitherto overlooked group of amines that those characterized here have been synthesized. By direct comparison with the isomeric α-methyl homologs synthesized by one of us[4] the effect of moving the alkyl group from the α- to the β-carbon may be ascertained. As might be anticipated a comparison of the physical properties shows only a slight change with this change in structure. Pharmacological work now in progress indicates, however, a much more radical change in activity than might be expected. Toxicities of the β-methyl series are less than for the corresponding α-methylamine.

Experimental

The preparation of the β-methyl-β-phenethylamines follows the general outline for the preparation of the α-

(1) Kalamazoo College Fellow, 1936–1937.

(2) These data are from a thesis submitted by Earl Pierson as a part of the requirements for the degree of Master of Science from Kalamazoo College, June, 1937.

(3) Barger and Dale, *J. Physiol.*, **51**, 19 (1910).

(4) Woodruff and Conger, THIS JOURNAL, **60**, 465 (1938). References to extensive reviews are given in this article.

(5a) Hartung and Munch, *ibid.*, **53**, 1879 (1931).

(5b) Tainter, *Arch. Internat. Pharm. and Therap.*, **46**, 205 (1933).

(6) Freund and König, *Ber.*, **26**, 2875 (1893).

(7) Von Braun, Grabowski and Kirschbaum, *ibid.*, **46**, 1280 (1913).