colorless needles, m.p. 120–121°. The compound gave a correct analysis for $C_{20}H_{32}O_2$.[7]

17β-Hydroxy-5β-androstan-3-one. Twenty-nine grams of testosterone was dissolved in 290 ml. of absolute alcohol containing 290 mg. of palladium black and 7.25 g. of potassium hydroxide previously dissolved in 15 ml. of distilled water, and hydrogenated at an initial pressure of 45 lb. for 2 hr. The work-up of the hydrogenation mixture was the same as described above, except that purification of the dried crude product was accomplished by dissolving in 500 ml. of a solution of benzene and ether 2:1, passing the solution through a chromatographic column containing 150 g. of chromatographic grade alumina (Harshaw), and eluting with 3 l. of a solution of benzene–ether 2:1. Evaporation of the eluate to dryness and crystallization from 500 ml. of hot petroleum ether (b.p. 60–110°) afforded 17.3 g. (60%) of pure 17β-hydroxy-5β-androstan-3-one, colorless plates, m.p. 142–144° (Kofler stage), $[\alpha]_D^{21}$ +32° (ethanol). Its infrared spectra was identical with that for an authentic sample of 17β-hydroxy-5β-androstan-3-one, and the mixed m.p. showed no depression.

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(7) We are greatly indebted to the Cancer Preparations Laboratory of Merck Sharp and Dohme for determination of optical rotation and carbon-hydrogen analyses of the compound.

Reaction of Diosgenin Acetate with Hydrogen Chloride in Acetic Anhydride

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In 1956 Miner and Wallis described an interesting fission of the spiroketal ring juncture of diosgenin acetate (I) with anhydrous hydrogen chloride in refluxing acetic anhydride to give an acetoxy chloro derivative.[1] The chloro compound, which crystallizes remarkably well from the resinous reaction product in 10% yield, was tentatively considered by its discoverers to be 3β,27-diacetoxy-16α-chloro-25α-5-cholesten-22-one. An attempt to use the substance as starting material for synthesis of steroid pyrroline derivatives of a type recently prepared,[2] however, has established that it must possess the structure 3β,16β-diacetoxy-27-chloro-25α-5-cholesten-22-one (II).

(1) R. S. Miner, Jr., and E. S. Wallis, *J. Org. Chem.*, 21, 715 (1956).

(2) F. C. Uhle and F. Sallmann, *J. Am. Chem. Soc.*, 82, 1190 (1960).

Treatment of II with three equivalents of potassium phthalimide in dimethylformamide at 105° gave the phthalimido derivative III. Hydrolysis of III with 5% ethanolic potassium hydroxide, followed by phthalamidic acid ring closure with *N*-cyclohexyl-*N'*-[2-(4-morpholinyl)ethyl]carbodiimide metho-*p*-toluenesulfonate, afforded the hemiketal IV, an intermediate in the synthesis of solasodine (VII) from kryptogenin.[3] Hemiketal dehydration of IV with glacial acetic acid at 100° gave the phthalimido furostene V, an intermediate in the synthesis of solasodine (VII) from pseudodiosgenin.[3]

Hydrazinolysis of III afforded VI, a tetrahydropyridine derivative first prepared from solasodine (VII) with acetic anhydride in the presence of zinc chloride.[4] Subjection of the gross product from hydrazine treatment of III to acetate hydrolysis with aqueous ethanolic potassium hydroxide gave solasodine (VII), constituting a three-step synthesis of the alkaloid from the sapogenin.

EXPERIMENTAL[5]

3β,16β-Diacetoxy-27-chloro-25α-5-cholesten-22-one (II). The compound ($C_{31}H_{47}ClO_5$) (535.14) was prepared from diosgenin acetate with anhydrous hydrogen chloride and refluxing acetic anhydride according to the procedure of Miner and Wallis.[1]

3β,16β-Diacetoxy-27-phthalimido-25α-5-cholesten-22-one (III). A magnetically stirred solution of 267 mg. (0.0005 mole) of 3β,16β-diacetoxy-27-chloro-25α-5-cholesten-22-one (II) and 277 mg. (0.0015 mole) of potassium phthalimide in 2 ml. of anhydrous dimethylformamide was heated at 105° for 24 hr.[6] After the mixture had been diluted with 20 ml. of saturated aqueous potassium chloride, the precipitate was collected by filtration, washed with water and dried. Initial attempts to recrystallize the product from methanol were erratic, giving low melting material which did not improve and even appeared to deteriorate on repetition, suggesting an instability to prolonged heating in alcoholic solution. Recrystallization was accomplished by brief warming in isopropyl alcohol, followed by rapid chilling. Two such recrystallizations gave 200 mg. (62%) of very small plates, m.p. 160–180°. The analytical sample, from isopropyl alcohol, melted at 179–182°; $[\alpha]_D$ + 14° (chloroform); infrared spectrum (KBr): 5.80 (acetoxy), 5.65, 5.90, 13.8, 14.0 μ (phthalimido).

Anal. Calcd. for $C_{39}H_{51}NO_7$ (645.81): C, 72.43; H, 7.96; N, 2.17. Found: C, 72.25; H, 7.82; N, 2.43.

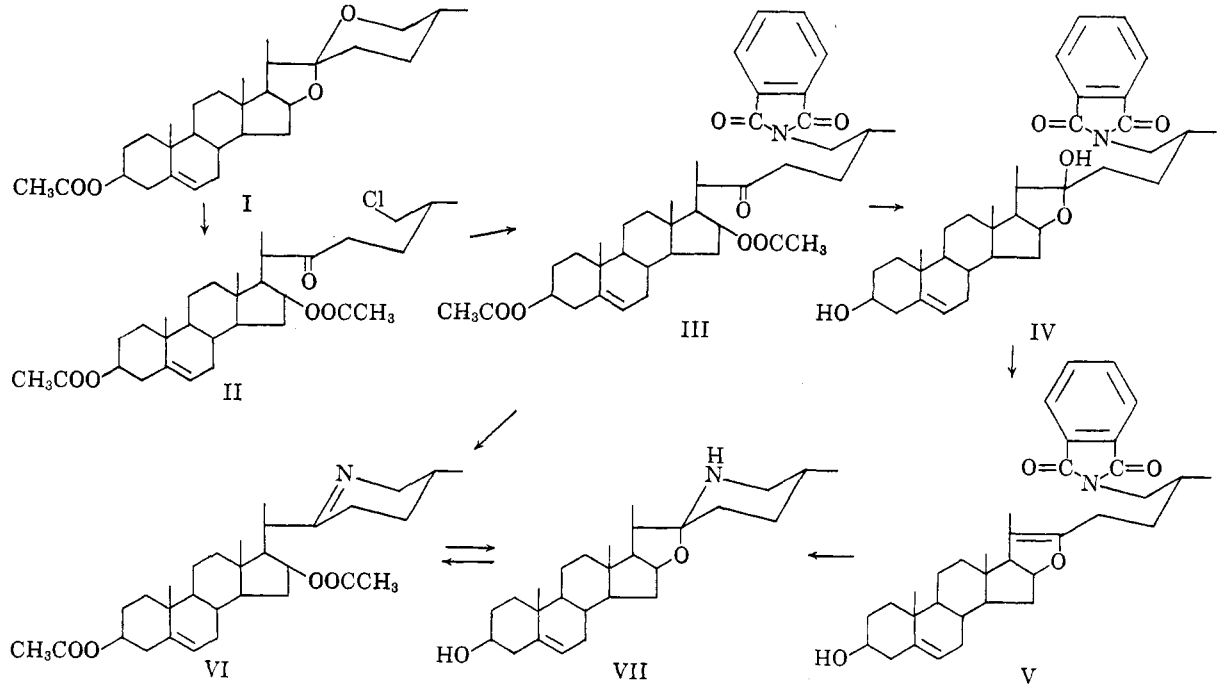
3β,22-Dihydroxy-27-phthalimido-25α-5-furostene ($C_{35}H_{47}NO_5$) (561.73) (IV). A solution of 129 mg. (0.0002 mole)

(3) F. C. Uhle, *J. Am. Chem. Soc.*, 83, 1460 (1961).

(4) Y. Sato, H. G. Latham, Jr., and E. Mosettig, *J. Org. Chem.*, 22, 1496 (1957); Y. Sato and N. Ikekawa, *J. Org. Chem.*, 25, 786 (1960).

(5) Melting points were observed on a calibrated micro hot stage. Microanalyses were performed by Dr. S. M. Nagy, Massachusetts Institute of Technology, Cambridge, Mass. Rotations were measured by Schwarzkopf Microanalytical Laboratories, Woodside 77, N. Y. Infrared spectra were recorded with a Perkin-Elmer spectrophotometer, model 137. Only those functional bands of significance in interpretation are mentioned.

(6) The sparingly soluble 27-chloro compound was altogether unreactive with potassium phthalimide in dimethylformamide at 25° under conditions used with 27-iodo derivatives.[3]



of 3β,16β-diacetoxy-27-phthalimido-25α-5-cholesten-22-one (III), 112 mg. (0.002 mole) of potassium hydroxide and 1 drop of water in 2 ml. of absolute ethanol was heated under reflux for 3 hr. After the mixture had been diluted with 10 ml. of water, the clear solution was acidified with 6*N* aqueous hydrochloric acid. The precipitate was collected by filtration, washed with water, and dried. To a solution of this phthalamidic acid in 1 ml. of methanol was added 212 mg. (0.0005 mole) of *N*-cyclohexyl-*N'*-[2-(4-morpholinyl)-ethyl] carbodiimide metho-*p*-toluenesulfonate.[7] After 3 hr. at 25°, followed by 20 hr. at 0°, the precipitate was collected by filtration and was washed with methanol. The filtrate was diluted with water to give a precipitate which was collected by filtration, washed with water, dried and combined with the material which had deposited directly from the methanolic reaction mixture. Two recrystallizations from methanol gave 56 mg. (50%); m.p. 170–173° (varies somewhat with rate of heating); rich infrared spectrum (KBr) identical with that of the product prepared by sodium borohydride reduction of 3β-hydroxy-27-phthalimido-25α-5-cholesten-16,22-dione: 5.65, 5.90, 13.8, 14.0 μ (phthalimido).[3]

Although phthalamidic acid ring closure of the sodium borohydride reduction product of 3β-hydroxy-27-phthalimido-25α-5-cholesten-16,22-dione with *N*-ethyl-*N'*-[2-(4-morpholinyl)ethyl]carbodiimide metho-*p*-toluenesulfonate at 25° had been accompanied by hemiketal dehydration,[3] IV was stable to treatment with as much as 20 equivalents of *N*-cyclohexyl-*N'*-[2-(4-morpholinyl)ethyl]carbodiimide metho-*p*-toluenesulfonate in methanol for 20 hr. at 25°.

3β-Hydroxy-27-phthalimido-25α-5,20(22)-furostadiene ($C_{35}H_{45}NO_4$) (*543.72*) (V). A solution of 20 mg. (0.000035 mole) of 3β,22-dihydroxy-27-phthalimido-25α-5-furostene (IV) in 12 drops of glacial acetic acid was heated at 100° for 10 min.[8] After the solution had been diluted with 10 ml. of water, the precipitate was collected by filtration, washed with water, and dried. Recrystallization from isopropyl alcohol gave 10 mg. (50%); m.p. 70–72°, followed by solidification and re-melting at 130–145°.

Samples of V, prepared from kryptogenin[3] and from pseudodiosgenin,[3] which had been stored for over a year no longer exhibited the initial low melting point (70–72°). Fresh recrystallization from isopropyl alcohol, however, gave material which melted at 70–72°, followed by solidification in characteristic fashion and re-melting at 130–145°. This material, when mixed with the substance prepared from III, again melted at 70–72°/130–145°. Infrared spectra (KBr) of the samples from the three sources were identical: 5.92 (medium) (shoulder) (vinyl ether), 5.70, 5.90, 13.8, 14.0 μ (phthalimido).

3β,16β-Diacetoxy-22(27)-imino-25α-5,22(N)-cholestadiene ($C_{31}H_{47}NO_4$) (*497.69*) (VI). A solution of 129 mg. (0.0002 mole of 3β,16β-diacetoxy-27-phthalimido-25α-5-cholesten-22-one (III), 64 mg. (0.002 mole) of hydrazine, and 1 ml. of methanol in 1 ml. of dichloromethane was kept at 25° for 3 hr.; a heavy deposit, presumably the hydrazine salt of phthalhydrazide, had begun to separate. After 68 hours at 0°, the mixture was diluted with water and was extracted with ether. The organic phase was washed with dilute aqueous ammonia and with water and was concentrated under reduced pressure. The residue was triturated with 10 ml. of 10% aqueous acetic acid. After a very small amount of insoluble material had been removed by filtration through cotton, the filtrate was made basic with dilute aqueous ammonia. An ether extract of the precipitate was washed with water, dried over anhydrous magnesium sulfate, and filtered. The residue from vacuum evaporation of the filtrate was recrystallized from methanol to give 55 mg. (55%) of plates; m.p. 184–192°; $[\alpha]_D$ +43° (chloroform); infrared spectrum (KBr): 5.80 (acetoxy), 6.05 μ (medium) (C=N).[4]

Gentle alkaline hydrolysis of VI gave a product still containing an acetoxyl residue, presumably at C-16,[4] as attested by infrared spectrum; more vigorous basic hydrolysis gave solasodine (VII),[4] identified by melting point and infrared spectrum.

Solasodine [*3β-Hydroxy-22(27)-imino-25α-5-furostene*] ($C_{27}H_{43}NO_2$) (*413.62*) (VII) A solution of 129 mg. (0.0002 mole) of 3β,16β-diacetoxy-27-phthalimido-25α-5-cholesten-22-one (III), 64 mg. (0.002 mole) of hydrazine and 1 ml. of methanol in 1 ml. of dichloromethane was kept at 25° for 45 hr. After the mixture had been diluted with water, the organic solvents were distilled under diminished pressure. The precipitate was collected by filtration and was washed with dilute aqueous ammonia and with water. To a solution of the precipitate in 4.5 ml. of ethanol was added a solution of 224 mg. (0.004 mole) of potassium hydroxide in 0.5 ml. of water.

(7) Aldrich Chemical Company, Milwaukee, Wis.

(8) For acetic acid dehydration of 22-hemiketals, *cf.* H. Hirschman and F. B. Hirschman, *Tetrahedron*, **3**, 234 (1958); Y. Sato and N. Ikekawa, *J. Org. Chem.*, **25**, 789 (1960).

After 20 hr. at reflux temperature, the solution was concentrated under reduced pressure. The residue was diluted with water and was extracted with ether. The organic phase was washed with water, dried over anhydrous magnesium sulfate, filtered, and concentrated *in vacuo*. One recrystallization of the residue from methanol gave 25 mg. (30%) of white lustrous plates, m.p. 199–201° (plates in part characteristically rearrange to needles as the melting point is approached; melt solidifies to long needles); melting point of a mixture with a sample of naturally occurring solasodine (m.p. 199–201°) 199–201°; infrared spectrum (KBr) identical with that given by a specimen of natural solasodine: 10.3, 10.4, 11.2, 11.5 μ (azaoxaspirane bands).

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Synthetic Polysaccharides. VII. Preparation of Polyglucose Nitrate

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In order to prepare water soluble cationic derivatives of the synthetic polysaccharides for macromolecular interaction studies and for biochemical applications,[1] various reactions were recently investigated[2] for the introduction of amine groups into the synthetic polyglucose.[3] One of these reactions was the attempted reduction of polyglucose nitrate with sodamide in liquid ammonia. Unfortunately, the reduction attempt gave only highly degraded unidentified products of low molecular weight. The preparation and the properties of the nitrate ester of the synthetic polyglucose are given, however, for the following reasons:

The chemically synthesized polymer of glucose is a highly branched polysaccharide,[3] having on the average three free hydroxyls available for esterification per anhydroglucose unit. A polymer with high nitrate content has a large number of nitrate groups in a very small space, held together by the covalently linked, branched, spherical carbohydrate skeleton:[4] In this regard it was interesting to note that a polyglucose dinitrate detonated with one half of the impact force necessary for nitrocellulose of similar degree of substitution; also it ignited at lower temperature (155°). These observations indicate that the close proximity of the nitrate groups attached to different monosaccharide residues also increases the instability of the polysaccharide nitrates as does the degree of substitution or the ratio of nitrogen to carbon and oxygen.

Undoubtedly this method of nitration of polyglucose can be applied to the numerous different synthetic polysaccharides which were prepared by similar polycondensation of various other carbohydrates.[5]

EXPERIMENTAL

Concentrated nitric acid (70% reagent grade; sp. gr. 1.42) 400 ml. and 500 ml. of concentrated sulfuric acid (96% reagent grade; sp. gr. 1.84) were mixed in a round bottomed flask, 2 l. capacity, equipped with a glass stirrer and a thermometer. The mixture was then chilled (ice-salt bath) to −3° to −4° and 20 g. (0.123 anhydroglucose unit) of finely divided polyglucose (number average molecular weight $\overline{M}_n$ = 6,600, intrinsic viscosity $[\eta]$ = 0.05, sample A, Ref. 2) was added as rapidly as possible with stirring. The temperature of the reaction mixture rose to −2° for several minutes then dropped to −3° where it was maintained for 90 min. with continued stirring. The ice bath was replaced with a warm water bath and the mixture was heated gradually over a 20-min. period to 34 ± 1°, maintaining this temperature for 10 min. The mixture was then poured onto 2000 g. of crushed ice plus 300 ml. of water. After the ice had melted the white lumpy product was separated from the aqueous phase by centrifugation. In order to free the ester from acidic material the former was exhaustively washed with water, 5% sodium bicarbonate, and finally with water again until neutral to litmus, separating it from the washings each time by centrifugation. The product, after storing under water in the refrigerator (+3°) overnight, became slightly acidic again. Next, in order to preclude the possibility that acidic material was being entrapped in the slightly granular state of the ester, the latter was carefully ground to a fine powder under water with an all glass mortar. The slurry was transferred to a sheet of filter paper (Whatman #54) in a Buchner funnel and again washed with water until free of acidic material. After being dried for about a week in a vacuum (0.1 mm.) desiccator over calcium chloride (anhyd.) and sodium hydroxide pellets, the polyglucose nitrate was obtained as a fine white powder with a slight but still persistent odor of nitrogen oxides. The nitrate was quite soluble in acetone, abs. ethanol, 95% ethanol–abs. ether mixture (1:1); fairly soluble in 95% ethanol; very slightly soluble in abs. ether; and insoluble in water. On continued periods of storage in the dry state the nitrate was observed to give off increasing amounts of the yellow oxides of nitrogen when shaken or stirred, and it finally became lemon-yellow in color. It was then concluded that the persistence of the acidity in the nitrate was due to some auto-catalytic decomposition of the nitrate linkages in the ester structure, possibly initiated by presence of small amount of sulfuric acid esters[6] and not to any mechanical entrainment of nitric acid in the granular or powdered material. From then on it was decided that for periods of storage longer than two to three days the poly-

(1) *Cf.* for example P. T. Mora and B. G. Young, *Arch. Biochem. Biophys.*, **82**, 6 (1959); P. T. Mora, B. G. Young, and M. J. Shear, *Makromol. Chemie*, **38**, 212 (1960); and B. G. Young and P. T. Mora, *Virology*, **12**, 493 (1960).

(2) J. W. Wood and P. T. Mora, in preparation.

(3) P. T. Mora and J. W. Wood, *J. Am. Chem. Soc.*, **80**, 685 (1958), P. T. Mora, J. W. Wood, P. Maury, and B. G. Young, *J. Am. Chem. Soc.*, **80**, 693 (1958).

(4) P. T. Mora, *J. Polymer Sci.*, **23**, 345 (1957).

(5) P. T. Mora and J. W. Wood, *J. Am. Chem. Soc.*, **82**, 3418 (1960).

(6) R. W. Kerr in *Chemistry and Industry of Starch*, Academic Press, Inc., New York, N. Y., 2nd Ed. 1950, p. 303, mentions this possibility in the case of the instability of starch nitrates.