recrystallized from 95% ethanol. Upon melting, the compound resolidified and then was charred at about 205°.

Anal. Calc'd for C_9H_7NO: C, 74.44; H, 4.87; N, 9.65. Found: C, 74.45; H, 4.82; N, 9.26.

Absorption in the infrared at 2.93 μ as well as a positive ferric chloride test suggested the presence of a phenolic hydroxyl group.

Preparation of 3-isoquinolylurea. The method of Kurzer[11] was followed. A solution of 1.4 g. (9.7 mmoles) of 3-aminoisoquinoline, 6 ml. of glacial acetic acid, and 10 ml. of water was warmed to 35–45°. A solution of 1.6 g. (9.7 mmoles) of potassium isocyanate in 10 ml. of water was warmed to 35–45° and added dropwise to the amine solution until a cloudiness appeared. The remainder of the isocyanate solution then was added all at once. The mixture was allowed to stand at room temperature for 2 hours and then was chilled in the refrigerator overnight. A light yellow precipitate of 3-isoquinolyl urea was collected and recrystallized from aqueous ethanol from which it separated as a white powder 1.0 g. (56%), m.p. 207–210°.

Anal. Calc'd for $C_{10}H_9N_3O$: N, 22.45. Found: N, 22.47.

Attempted nitrosation of 3-isoquinolylurea. A solution of 0.5 g. (2.7 mmoles) of 3-isoquinolylurea, 3.2 g. of concentrated sulfuric acid, and 5 ml. of water was cooled to 0–5°. The dropwise addition of a solution of 2.8 g. (4.1 mmoles) of sodium nitrite in 6 ml. of water was accompanied with vigorous gas evolution. The mixture was stirred for an additional 1.5 hours. A yellow-orange precipitate, 0.24 g., was collected, recrystallized from aqueous ethanol from which it separated as a yellow powder, m.p. 162–164°, and dried in the vacuum oven at 50° for 24 hours. Elementary analyses suggested the empirical formula, $C_{20}H_{18}N_6O_5$ for this unidentified product. The compound gave a positive Liebermann's nitroso test.

Anal. Calc'd for $C_{20}H_{18}N_6O_5$: C, 56.85; H, 4.30; N, 19.90. Found: C, 57.40; H, 4.37; N, 20.09.

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An Improved Method of Preparation of Side-Chain Labeled Cholesterol

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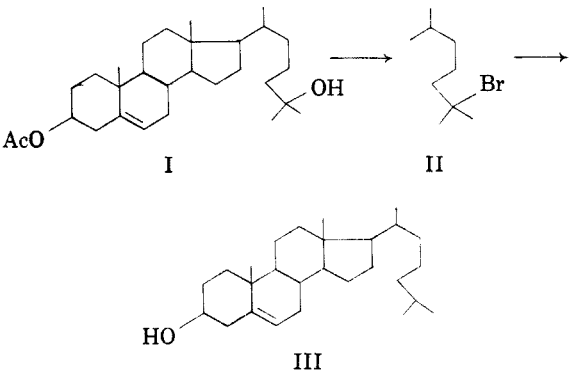
The conversion of 25-hydroxycholesteryl acetate[1,2] (I) to cholesterol (III) is of interest since the former compound is a readily prepared intermediate in the synthesis of cholesterol labeled with C^{14} at positions 24, 25, or 26. To date, two methods[1,2] have been reported for this conversion. In both instances, the 25-ol was first dehydrated to the 25-dehydro compound which, in turn, was either selectively hydrogenated directly or first converted to the 3,5-cyclo-6-ether and then hydrogenated. It has now been found that 25-bromocholesteryl acetate (II), an intermediate in the conversion of the 25-ol to the 25-dehydro compound, can be hydrogenated directly to cholesterol in alkaline solution in the presence of a moderately active Raney nickel catalyst. The over-all yield for this two-step conversion is 65%.



I II III

The intermediate 25-hydroxycholesteryl acetate was prepared by allowing methylmagnesium iodide to react with methyl 3β-hydroxy-Δ^5-homocholenate. In this manner, cholesterol-24-C^{14} was prepared by utilizing 3β-hydroxy-Δ^5-homocholenic acid-24-C^{14} which had been synthesized by chain elongation of the respective cholenic acid with C^{14}-diazomethane.[3]

EXPERIMENTAL[4]

Conversion of 25-hydroxycholesteryl acetate to cholesterol. The crystalline monoacetate[2] (490 mg.) was converted with phosphorus tribromide to 25-bromocholesteryl acetate as described previously,[2] and the bromide was recrystallized from acetone-water and dried in a high vacuum at room temperature; yield, 430 mg. (78%), m.p. 113–115° (lit.[2] m.p. 113.5–115.0°).

The recrystallized bromide (430 mg.) was dissolved in 80 ml. of 3.5% methanolic sodium hydroxide solution and was hydrogenated at a pressure slightly above atmospheric with a moderately active Raney nickel catalyst which had been prehydrogenated. Within 10–20 minutes, the theoretical amount of hydrogen was absorbed and then the catalyst was removed by filtration and washed with methanol. Potassium hydroxide (6.0 g.) was added to the filtrate and the solution was heated under reflux for 3 hours. The solution was concentrated under reduced pressure, the residue was cooled in ice, and a small volume of ether was added. The mixture was acidified with 1:1 hydrochloric acid, and the ethereal layer was separated, washed with water, and dried. After removal of the solvent, the residue was chromatographed on 12 g. of neutral alumina. With benzene-ether (1:1), 28 mg. (65%) of a crystalline material, m.p. 138–140°, was eluted. After recrystallization from methanol, the cholesterol melts from 145–146° and has an $[\alpha]_D^{25}$ −39.4° (*c*, 1.08, $CHCl_3$). The infrared spectrum was identical with that of an authentic sample.

Preparation of catalyst. The following method of Plattner and Pataki[5] was used. Raney nickel alloy (5 g.) was suspended in 20 parts of 4% aqueous sodium hydroxide solution and heated for 30 minutes on a steam-bath. This procedure was repeated with fresh sodium hydroxide solution

(1) A. I. Ryer, W. H. Gebert and N. M. Murrill, *J. Am. Chem. Soc.*, **72**, 4247 (1950).

(2) W. G. Dauben and H. L. Bradlow, *J. Am. Chem. Soc.*, **72**, 4248 (1950).

(3) The C^{14}-methylamine utilized in the preparation of labeled N-nitrosomethylurea was kindly supplied by Dr. K. H. Takemura.

(4) All melting points are corrected.

(5) Pl. A. Plattner and J. Pataki, *Helv. Chim. Acta*, **26**, 1241 (1943).

and the catalyst (3 g.) then was washed by decantation with water 20–30 times, then three times with absolute methanol, and stored under absolute methanol in the ice-box for not longer than two weeks.

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Long-Acting Androgens[1]

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Esterification of the hydroxyl groups has yielded derivatives of steroid hormones of desirable clinical utility which require less frequent administration for replacement therapy in hormone deficiency states. More important than the convenience and economy of such therapeutic measures is the release of the required replacement steroid at a physiological rate that does not invoke the potential dangers of overdosage and side reactions associated with derivatives which are absorbed rapidly and administered frequently.

Recent work with esters of testosterone[2–8] has yielded a variety of long-acting androgens far surpassing testosterone propionate (TP). Although the mode of action of these esters is still unsettled, it is believed that esterified steroids achieve their longer action because of slower absorption from the injection site.[7] Free testosterone,[9,10] however, appears to be required for the androgenic response.

If the sterol is varied, and the same acylating agent such as the hindered pivalic acid is used, then prolonged activity[11–13] or loss of activity[14] may result.

Our investigation of esters of testosterone and androstan-17-β-ol-on (Table I) involved relatively bulky acyl groups[2–8] and sterically hindered acyl groups[11–14] in an attempted conciliation of the factors of absorption and saponification to yield the free androgen at a rate compatible with physiological requirements.

The failure to obtain any androgenic response with the *tert*-butyl acetates and α,α-dibenzylacetates could be interpreted as due to resistance of such sterically hindered esters to saponification.[15–20]

With β-halopropionates, prolonged activity far superior to testosterone propionate or α-bromopropionate[21] was noted. The response with halogen in the *beta* position suggested that these structures might suffer metabolic dehydrohalogenation to yield testosterone acrylate. Synthesis and androgenic evaluation of testosterone acrylate showed activity considerably inferior to TP.

Testosterone β-chlorocrotonate was only mildly androgenic, paralleling observations[21] with the crotonate. The low androgenic potency of the α,β-unsaturated esters may be due to rapidity of their hydrolysis.[22]

An alternative possibility where the β-halopropionate yields acrylate which spontaneously polymerized *in vivo* was explored by polymerizing testosterone acrylate. The resultant polymer was so insoluble that it showed only slight androgenic activity.

Synthesis and evaluation of the group of testosterone esters such as the β-N-morpholinopropionate, β-N-pyrrolidinopropionate, hemisuccinate, and methyl ester of the hemisuccinate, indicated these

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(22) Meyers, Collett, and Lazell, *J. Phys. Chem.*, **56**, 461 (1952) report that an unsaturated carbon-to-carbon linkage near the carbonyl carbon in carboxylic esters always speeds the rate of hydrolysis compared to that of the saturated ester.