hydrous Terramycin in 400 ml. of dried dioxane at 10°. The solution was allowed to warm to room temperature. After 1.5 hours, nitrogen evolution had virtually ceased and the product, which had partially separated, was precipitated by the addition of 1400 ml. of commercial hexane. The amorphous product was dried *in vacuo* (16.9 g.) and then stirred with 100 ml. of methanol for 30 minutes. The crystalline insoluble precipitate (4.0 g.) which formed was further purified by recrystallization from 50% aqueous methanol to yield 2.0 g. (10% yield) of dimethylterramycin. This product decomposes without melting at about 225°. It is insoluble in water, pyridine and the common organic solvents.

Anal. Calcd. for $C_{24}H_{28}N_2O_9$: C, 59.00; H, 5.77; N, 5.73; OCH_3 (2), 12.69. Found: C, 59.23; H, 5.90; N, 5.69; OCH_3, 12.81.

The ultraviolet absorption spectrum is similar to that of Terramycin, λ_{max} 272 mμ, log ε 4.40 and λ_{max} 352 mμ, log ε 4.0.

The **hydrochloride** of dimethylterramycin was prepared by dissolution of the base in methanolic hydrogen chloride, precipitation with ether, and careful recrystallization from ethyl acetate–methanol containing hydrogen chloride; yellow hexagonal plates. The product decomposes without melting at 175° and has $[\alpha]^{26}D$ −110° (methanol). Titration in aqueous dimethylformamide showed pK_a 7.7, equivalent weight 540 (calcd. 525).

Along with the dimethylterramycin, there is obtained as the principal product (70% yield) an amorphous, unstable material which evolves trimethylamine on mild alkali treatment.

Diacetylterramycin (5,12a-Diacetoxy-4-dimethylamino-1,4,4a,5,5a,6,11,12a-octahydro-3,6,10,12-tetrahydroxy-6-methyl-1,11-dioxo-2-naphthacenecarboxamide).[45]—Ten grams of anhydrous Terramycin was dissolved in 200 ml. of anhydrous dioxane, and the solution made up to 1 l. with acetic anhydride. After 14 days at 25–30°, the solution was concentrated to dryness *in vacuo*, below 35°, and the crystalline product twice recrystallized from toluene to yield 8.8 g. (75%) of pure diacetylterramycin.

Anal. Calcd. for $C_{26}H_{28}N_2O_{11}$: C, 57.35; H, 5.18; N, 5.15; acetyl (2), 15.81; mol. wt., 544.5. Found: C, 57.59; H, 5.23; N, 5.11; acetyl, 15.69; equiv. wt., 546.

This product melts with decomposition at 208–213° and has $[\alpha]^{25}D$ +211° (acetone). Titration in dimethylformamide–water shows pK_a 6.75 and 8.85. Terramycin can be regenerated from this product by the action of 1 *N* aqueous sodium hydroxide at 25° for 5 minutes.

Benzenesulfonylterramycinonitrile (10-Benzenesulfonoxy-4-dimethylamino-1,4,4a,5,5a,6,11,12a-octahydro-3,5,6,12,-12a-pentahydroxy-6-methyl-1,11-dioxo-2-naphthacenenitrile).[46]—Benzenesulfonyl chloride, 3.6 g., was added to a solution of 2.5 g. of Terramycin hydrochloride in 7 ml. of pyridine at 5° and held at 5° overnight. The solution was poured into 50 ml. of ether and the gummy solid stirred for 1 hour with 25 ml. of water to yield 2.6 g. of a light tan crystalline product which was purified by two recrystallizations from dimethylformamide, washed with acetone and dried *in vacuo* at 100° for 3 hours. This product, m.p. 210–211°, contains a molecule of dimethylformamide of crystallization.

Anal. Calcd. for $C_{28}H_{28}N_2O_{10}S \cdot C_3H_7NO$: C, 56.78; H, 5.07; N, 6.41; S, 4.89; mol. wt., 655.7. Found: C, 57.33; H, 5.32; N, 6.13; S, 5.44.

Titration shows an equivalent weight of 680, pK_a 6.95 $[\alpha]^{25}D$ −378° (dimethylformamide). The ultraviolet absorption spectrum is similar in shape to that of Terramycin, λ_{max} 275 mμ, log ε 4.23, and λ_{max} 342 mμ, log ε 4.06 in acid methanol. The infrared spectrum shows the characteristic nitrile absorption band at 4.5 μ and carboxamide absorption at 6.05 μ, due to dimethylformamide. The crystalline monohydrate of benzenesulfonylterramycinonitrile, prepared by suspending the dimethylformamide solvate in water, no longer shows strong absorption at 6.05 μ.

On acetylation in acetic anhydride–pyridine, this compound yields a **triacetate** which was recrystallized from ethanol and dried *in vacuo* at 100° for 5 hours; $[\alpha]^{25}D$ +8° (dimethylformamide), pK_a 5.3.

Anal. Calcd. for $C_{34}H_{32}N_2O_{13}S \cdot {}^1/_2H_2O \cdot {}^1/_2CH_3CH_2OH$: C, 56.75; H, 4.90; N, 3.78; acetyl, 17.43; ethoxyl, 3.04; water, 1.21; mol. wt., 740.7. Found: C, 56.90; H, 4.71; N, 4.10; acetyl, 17.12; ethoxyl, 2.84; water (Karl Fischer), 0.8; equiv. wt. (titration), 765.

Terranaphthol (3-Hydroxymethyl-4-methyl-1,8-naphthalenediol) (XVI).[47]—The isolation of this compound has been described.[5] Purification through the triacetate, m.p. 148.7–149.4°, yielded XVI as the pure compound, m.p. 172.4–173.0°. XVI gives a green color with alcoholic or aqueous ferric chloride and a red precipitate with aminoantipyrine.[48] Titration showed an equivalent weight of 207 (calcd. 204), pK_a 7.5. A Kuhn–Roth C-methyl determination showed 6.2% C-methyl (calcd. for 1 C-methyl, 7.35%).

The ultraviolet absorption spectrum shows a major peak, λ_{max} 232 mμ, log ε 4.79 and a weaker group of peaks, λ_{max} 312–341 mμ, log ε 3.89 (Fig. 3).

The oxidation of terranaphthol with peroxide, nitric acid or potassium dichromate in acetic acid yielded intractable tars. An attempt to oxidize terranaphthol with a suspension of silver oxide in ether yielded only unchanged starting material.

Terranaphthol Monomethyl Ether.—When a dioxane solution of terranaphthol was added to 5 moles of diazomethane in ether solution and held at 25° overnight, a monomethyl ether[49] could be isolated by distillation at 160° (0.05 mm.). This oil crystallized slowly from benzene–ligroin and from ether–ligroin to yield a product, m.p. 88–91°, in 25% yield. The infrared absorption spectrum in dioxane solution shows a single OH absorption band at 2.90 μ, in contrast to the two OH bands observed for terranaphthol. The ultraviolet absorption spectrum is virtually identical with that of the free phenol.

Anal. Calcd. for $C_{13}H_{14}O_3$: C, 71.54; H, 6.45; methoxyl, 14.21. Found: C, 71.68; H, 6.69; methoxyl, 14.38.

Effect of Terranaphthol on the Acidity of Boric Acid.—A solution of 40.8 mg. (0.2 mmole) of terranaphthol in 2 ml. of absolute ethanol was added to 5 ml. of 0.5 molar aqueous boric acid. The *p*H, as measured with a glass electrode instrument, was 2.20, while that of a blank prepared from 5 ml. of boric acid and 2 ml. of ethanol was 5.00. The observed *p*H change was, therefore, 2.8 *p*H units. This result, together with others obtained in a similar manner, is reported in Table I. It should be noted that the change in *p*H is due in part to the natural acidity of the added organic compound.

Terranaphthoic Acid (1,8-Dihydroxy-4-methyl-3-naphthoic Acid) (XXXII).—Two-tenths gram of terranaphthol was ground in a mortar with 2 g. of sodium hydroxide and 2 g. of potassium hydroxide, placed in a nickel crucible and immersed in a metal bath preheated to 260–270°. After 15 minutes, when the evolution of gas had nearly ceased, the brown melt was cooled, dissolved in 15 ml. of water, and quickly acidified with cooling to *p*H 1 using 4 *N* sulfuric

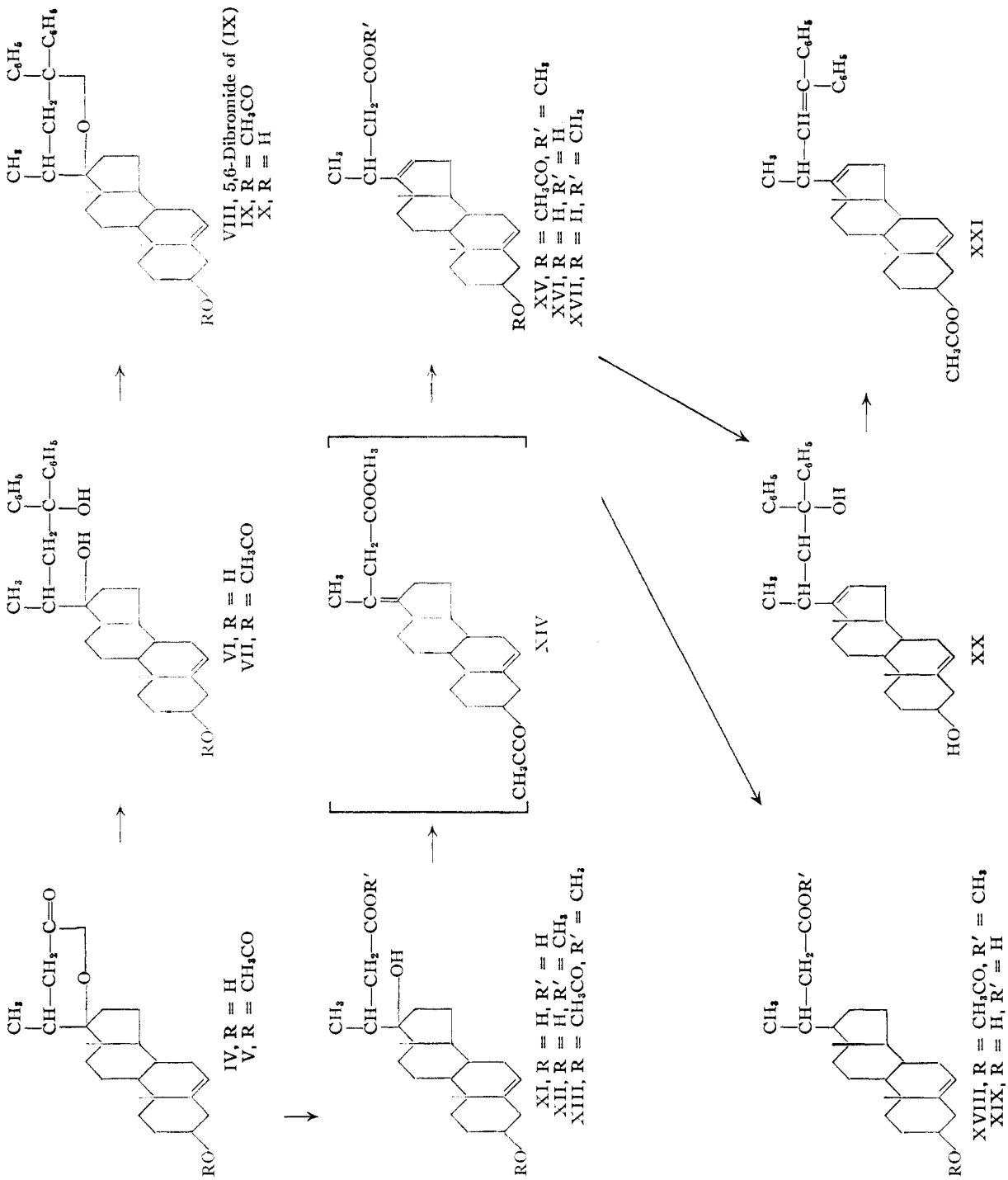
(45) The assignment of this structure is based on (i) the existence of two acid constants for this compound, (ii) the observation that it yields an anhydro compound analogous to anhydroterramycin chloroform–hydrogen chloride.

(46) (a) Consideration of the absorption spectra, the acid constants and chemical properties of this compound and its degradation products have led us to assign the benzene-sulfonyl group to the 10 position; (b) the conversion of amides to nitriles by the action of acid halides in pyridine has precedent, *cf.* Q. E. Thompson, THIS JOURNAL, **73**, 5841 (1951), and J. Mitchell Jr., and C. Ashby, *ibid.*, **67**, 161 (1945).

(47) The experiments on terranaphthol *per se* did not conclusively distinguish between XVI, and an alternative containing a $-CH_2OH$ at C-6. Thus, the group could not be placed at C-2 or C-7 for these reasons: (i) the acidity of terranaphthoic acid and its ultraviolet spectrum differ markedly from those of 1,8-dihydroxy-2-naphthoic acid; (ii) the carbonyl absorption of terranaphthoic acid occurs at 5.85 μ, as contrasted with that (6.05 μ) of 1,8-dihydroxy-2-naphthoic acid (salicylic acid type). Further, the positive aminoantipyrine test (*cf.* footnote 48) of both terranaphthol and terranaphthoic acid, and the relative difficulty of decarboxylation of the latter, as compared with 1,8-dihydroxy-4-naphthoic acid, exclude attachment at C-5. Of the remaining positions, C-3 and C-6, the former must be chosen, on the basis of the oxidations of methylated derivatives of terrinolide (XXXVII) and decarboxamidoterrinolide (XXXVIII) to xii and XXXIV.

(48) E. Emerson, H. Beacham and L. Beigle, *J. Org. Chem.*, **8**, 417 (1943).

(49) 1,8-Naphthalenediol, prepared by alkali fusion of 1-naphthol-8-sulfonic acid, likewise yields a monomethyl ether with diazomethane. See J. Böeseken and L. G. Smitt, *Rec. trav. chim.*, **58**, 125 (1939).



whose analysis showed the loss of one mole of water. The dibromo compound was unchanged when treated with ozone, indicating that the dehydration had not produced an olefinic bond; the debrominated product was also unchanged when refluxed with acetic anhydride or when heated with phosphorus oxychloride and pyridine. In addition, the lack of strong infrared absorption at 3μ[8] showed the absence of a hydroxyl group. From this evidence, it was concluded that the dehydrated compound was an ether (VIII). Attempts were made to dehydrate the 3-acetate triol (VII) to produce side chain unsaturation but only the ether (IX) could be obtained.

The lactone (IV) was also opened by refluxing with alcoholic potassium hydroxide, followed by careful acidification to give 3,17-dihydroxynor-5-cholenic acid (XI). The silver salt of this acid was converted to the corresponding methyl ester (XII) by treatment with methyl iodide. Attempts to esterify by heating with methanol in the presence of sulfuric acid or treatment with dimethyl sulfate resulted only in regeneration of the lactone (IV). Mild acetylation of the methyl ester (XII) in

(8) R. B. Barnes, R. C. Gose, R. W. Stafford and V. Z. Williams, *Anal Chem.*, **20**, 402 (1948).

pyridine gave the 3-acetate (XIII). The acetoxy lactone (V) was produced when the ester (XII) was refluxed with acetic anhydride. When treated with thionyl chloride, the acetoxy ester (XIII) was readily dehydrated to a diene. The lack of ultraviolet absorption at 230 mμ[9] showed that the new double bond introduced was not conjugated with the ester group, indicating a diene structure of either (XIV) or (XV). Reduction of the diene (XIV) or (XV) with palladium catalyst gave methyl, 3β-acetoxynor-5-cholenate (XVIII). This ester (XVIII) was conclusively identified by comparison with an authentic sample prepared from 3β-acetoxybisnor-5-cholenic acid by Arndt–Eistert homologation.[10]

In an attempt to locate the double bond produced by dehydration, the diene (XIV) or (XV) was treated with one equivalent of bromine, in the hope that the 5,6-bond would brominate preferentially, and the mixture was ozonized. No identifiable products were isolated.

The diene (XIV) and (XV) was refluxed with strong ethanolic potassium hydroxide for 48 hours using the procedure of Fittig,[11] who isomerized β,γ-unsaturated acids to their α,β-isomers by this technique. No double bond shift occurred; this was interpreted to indicate that the double bond was in position 16,17 as in (XV). This assumption was confirmed in the following manner: The diene was treated with phenylmagnesium bromide to give the corresponding diphenylcarbinol (XX). This product after dehydration gave a triene which exhibited strong absorption at 250 mμ (log ε 4.2), the expected result for a diphenylethylene like (XXI). If the diene had the structure (XIV), the dehydration should have produced a diphenylethylene conjugated with a 17,20-double bond causing a bathochromic shift of about 60 mμ and an expected absorption band at 310 mμ.[12,13]

To clarify the configuration at C_{17}, Fisher–Hirschfelder models were constructed. Both the 17α- and β-hydroxy lactones could be assembled readily. The diphenyl ether (X) model with the ether oxygen in the 17α-position has both phenyl groups at C_{23} in back of the D ring far removed from the angular methyl group at C_{13}; both phenyl groups have considerable freedom of rotation. Model (X) with the ether oxygen in the 17β-position is strongly hindered with one phenyl group in close proximity to the C_{13} angular group; movement of this phenyl group is sharply limited. It appears likely therefore that the ether oxygen at C_{17} is in the α-position. The ether (X) was produced by opening the lactone with Grignard reagent followed by very mild dehydration, neither of which reactions are expected to cause inversion, so that it was indicated that the oxygen atom at C_{17} in the lactone (V) probably also possesses the α configuration. Furthermore, the C_{17} side chain in naturally occurring steroids is believed to be in the β-position; the introduction of a hydroxyl group at C_{17} by oxidation to produce the lactone should occur without inversion in the α-position.

An examination of the models of both the lactone (V) and the ether (X) with 17α-oxygen shows that the C_{20} hydrogen and the C_{20} methyl group are both above the plane of the D ring, and that only one position in space is possible for the C_{20} methyl group, *i.e.*, attached at that portion of the C_{20} atom farthest removed from the C_{13} angular methyl group. This is shown diagrammatically in Fig. 1. In the diphenyl triol (VI) the side chain at C_{20} is therefore in the β-position according to the convention of Fieser and Fieser.[14] This agrees with the configuration which these investigators have suggested for the naturally occurring steroids based on optical rotational evidence.

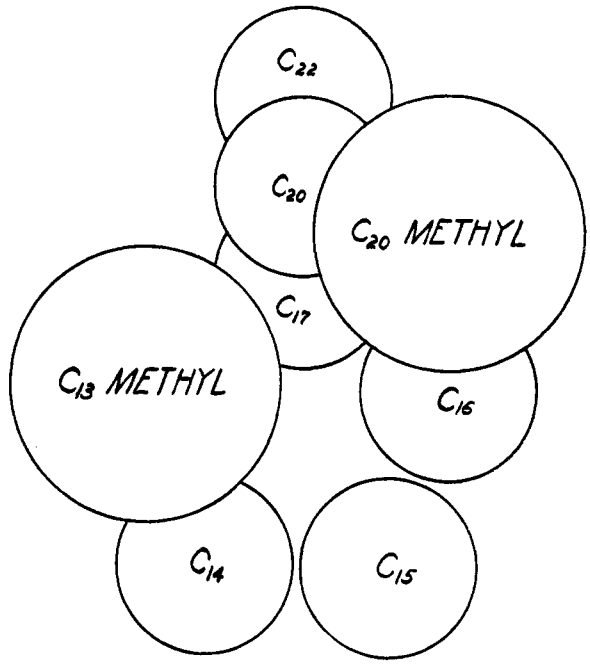


Fig. 1.—Front view of the D ring of the 17α-hydroxy lactone (V) drawn from a Fisher–Hirshfelder model.

Experimental[15]

Isolation of 3β,17α-Dihydroxynor-5-cholenic Acid Lactone (IV).—The mother liquor from a methanol recrystallization of crude dehydroepiandrosterone acetate obtained by the chromic acid oxidation of sitosterol acetate dibromide[16] was concentrated to dryness. To a solution of 61 g. of this residue in 720 cc. of hot methanol was added a solution of 30.5 g. of anhydrous potassium carbonate dissolved in 92 cc. of water. The mixture was refluxed for two hours, poured into water and made faintly acid to litmus with acetic acid. The precipitate was filtered, washed neutral with water and dried. The product was dissolved in hot ethyl acetate, concentrated to a volume of 400 cc. and cooled to 17°. The

(9) L. Ruzicka, Pl. A. Plattner and J. Pataki, *Helv. Chim. Acta*, **25**, 425 (1942).

(10) "Organic Reactions," Vol. 1, John Wiley and Sons, Inc., New York, N. Y., 1942, p. 38.

(11) R. Fittig, *Ann.*, **283**, 47 (1894).

(12) L. F. Fieser and M. Fieser, "Natural Products Related to Phenanthrene," A. C. S. Monograph No. 70, Third Ed., Reinhold Publ. Corp., New York, N. Y., 1949, p. 184.

(13) Ch. Meystre, A. Frey, A. Wettstein and K. Miescher, *Helv. Chim. Acta*, **27**, 1815 (1944), give the following data for the related compounds: Δ^{22}-3α,12β-diacetoxy-24,24-diphenylcholene, $\lambda_{max.}^{CHCl_3}$ 250 mμ, log ε 4.4; $\Delta^{20-22,22}$,3α,12β-diacetoxy-24,24-diphenylcholadiene, $\lambda_{max.}^{CHCl_3}$ 310 mμ, log ε 4.5.

(14) L. F. Fieser and M. Fieser, *Experientia*, **4**, 285 (1948); P. Wieland and K. Miescher, *Helv. Chim. Acta*, **32**, 1922 (1949).

(15) All melting points are corrected. Microanalyses and disagree microrotations by Edwin Conner of these laboratories.

(16) C. R. Addinall, Fiat Final Report 996, Technical Industrial Intelligence Division, U. S. Department of Commerce, 1947, p. 29.

crude hydroxy lactone was filtered and dried; yielding 6.5 g., m.p. 267–270°. The product was recrystallized from methanol to give 4.6 g. of fine needles melting at 282.2–283.4°; $[\alpha]^{22}D$ −94.4° (2% in $CHCl_3$).

Anal. Calcd. for $C_{23}H_{34}O_3$: C, 77.05; H, 9.56. Found: C, 76.97; H, 9.62.

This lactone readily formed the 5,6-dibromide when treated with one equivalent of bromine in acetic acid. The product when recrystallized from methanol melted at 127.2–127.4° (dec.). The unstable dibromide did not give a satisfactory analysis but when debrominated with sodium iodide in methanol by the method of Schoenheimer[17] regenerated the hydroxy lactone (IV).

3β-Acetoxy-17α-hydroxynor-5-cholenic Acid Lactone (V).—A solution of 3.3 g. of the hydroxy lactone (IV) in 60 cc. of dry pyridine and 10 cc. of acetic anhydride was warmed at 60° for one hour, allowed to stand overnight and then poured into water. The product was filtered, washed with water and dried, yielding 3.52 g., m.p. 203.5–207.5°. The acetate was recrystallized from methanol to give 3.3 g. of fine needles melting at 207.5–210.5°; $[\alpha]^{24}D$ −86.7° (2% in $CHCl_3$).

Anal. Calcd. for $C_{25}H_{36}O_4$: C, 74.96; H, 9.06. Found: C, 74.84; H, 9.01.

3β-Acetoxy-17α-hydroxy-5,6-dibromonor-5-cholenic Acid Lactone.—The acetoxy lactone (V) was treated with bromine in acetic acid and the dibromide crystallized from methanol to give fine needles melting at 158–159° (dec.); $[\alpha]^{22}D$ −66.0° (2% in $CHCl_3$). The acetate dibromide was unstable; the sharp odor of hydrobromic acid could be detected shortly after its preparation.

Anal. Calcd. for $C_{25}H_{36}O_4Br_2$: C, 53.58; H, 6.48; Br, 28.52. Found: C, 53.95; H, 7.20; Br, 27.70.

17α-Hydroxy-3-ketonor-4-cholenic Acid Lactone.—A mixture of 6.0 g. of the hydroxy lactone (IV), 260 cc. of dry toluene, 18 cc. of cyclohexanone and 3.1 g. of aluminum isopropoxide was refluxed for two hours. The reaction mixture was cooled, 50 cc. of water was added and then the mixture was steam distilled until no more oil passed over. Diatomaceous earth (25 g.) was added, the mixture filtered and the filter cake dried. The filter cake was extracted several times with methylene chloride and the pooled extracts concentrated to a small volume. The methylene chloride was completely replaced with ethyl ether by distillation, the solution concentrated to a thin slurry of crystals, cooled to 5° and filtered. The crude product (5.26 g.) was recrystallized from ethyl acetate to give 4.33 g. of needles, m.p. 210.6–212.2°; $[\alpha]^{24}D$ +52.2° (2% in $CHCl_3$); $\lambda_{max.}^{ethanol}$ 238 mμ, log ε 4.2.

Anal. Calcd. for $C_{23}H_{32}O_3$: C, 77.49; H, 9.05. Found: C, 77.35; H, 9.12.

3β,17α,23-Trihydroxy-23,23-dimethylnor-5-cholene.—A solution of 5.0 g. of the lactone acetate (V) in 100 cc. of anhydrous, thiophene-free benzene was added to a Grignard solution prepared from 7.0 g. of magnesium, 50 g. of freshly distilled methyl iodide and 300 cc. of anhydrous ethyl ether. The mixture was refluxed for three hours and allowed to stand overnight. After cooling to 5°, the complex was decomposed by the slow addition of 60 cc. of ice-water and 120 cc. of 50% acetic acid solution, and the mixture steam distilled until no more oil passed over. The product was filtered at room temperature, washed with water until neutral and dried at 50°, yielding 5.0 g., m.p. 159–174°. The crude triol was dissolved in hot methanol, treated with activated carbon and Adsorptive Magnesia Powder, No. 2642,[18] and filtered through a mat of diatomaceous earth. The filtrate was concentrated to a small volume and the methanol replaced with acetone until crystallization started to take place. After cooling to 5°, the crystals were filtered to give 1.35 g. of hexagonal plates, m.p. 235.5–237.4°; $[\alpha]^{24}D$ −85.9° (1% in pyridine).

Anal. Calcd. for $C_{25}H_{42}O_3$: C, 76.87; H, 10.84. Found: C, 76.71, H, 10.37.

3β,17α,23-Trihydroxy-23,23-diphenylnor-5-cholene (VI).—A solution of 38.0 g. of the lactone acetate (V) in a mixture of 700 cc. of anhydrous thiophene-free benzene and 240 cc. of anhydrous ethyl ether was added to a Grignard solution prepared from 24.3 g. of magnesium, 184 g. of bromobenzene and 320 cc. of anhydrous ethyl ether. The mixture was refluxed for two hours and allowed to stand overnight. After cooling to 5°, the complex was decomposed by the slow addition of 400 cc. of ice-water and 500 cc. of 20% acetic acid, and steam distilled until no more oil passed over. The somewhat oily product was filtered at 25° and washed thoroughly with water. The oils were removed by stirring with enough hot acetone to make a thin slurry, the mixture was then cooled to 5°, filtered and the crystals washed oil-free with cold acetone. The crude product (26.5 g.) was recrystallized from methanol to give 20 g. of needles, m.p. 268.0–270.0° with decomp.; $[\alpha]^{23}D$ −51.8° (2% in pyridine); $[\alpha]^{25}D$ −50.5° (1% in dioxane).

Anal. Calcd. for $C_{35}H_{46}O_3$: C, 81.67; H, 9.01. Found: C, 81.31; H, 8.89.

3β-Acetoxy-17α,23-dihydroxy-23,23-diphenylnor-5-cholene (VII).—A solution of 33 g. of the triol (VI) in 133 cc. of dry pyridine and 49 cc. of acetic anhydride was warmed at 60–70° for one hour, allowed to stand for three hours at room temperature and then poured into water. The product was filtered, washed neutral with water and dried, yielding 35.3 g., m.p. 236–241°. The product was recrystallized from methanol to give 33.5 g. of needles, m.p. 247.8–251.2°; $[\alpha]^{26}D$ −56.1° (2% in $CHCl_3$).

Anal. Calcd. for $C_{37}H_{48}O_4$: C, 79.81; H, 8.69; saponification number calculated for a monoacetate, 100.6. Found: C, 79.79; H, 8.66; sapn. equiv., 100.

3β-Acetoxy-5,6-dibromo-17,23-oxido-23,23-diphenylnor-5-cholene (VIII).—To a solution of 3.0 g. of the triol acetate (VII) in 200 cc. of C.P. chloroform was added at room temperature 0.865 g. of bromine dissolved in 25 cc. of chloroform over a five-minute period. The mixture was allowed to stand for one hour at room temperature and then concentrated on a steam-bath with a stream of air blowing over the surface until a heavy slurry of crystals was formed. Ethyl ether (85 cc.) was added, the mixture held at 5° for one hour and filtered. The crude product (2.0 g.) melting at 187–189° with decomp. was dissolved in chloroform, concentrated to a small volume and the chloroform completely replaced by methanol. The methanol solution was concentrated to a thin slurry of crystals, cooled to 5° and filtered to give 1.42 g. of purified dibromide, m.p. 188.9–190.6° with decomp.; $[\alpha]^{21}D$ −78.8° (2% in $CHCl_3$).

Anal. Calcd. for $C_{37}H_{46}O_3Br_2$: C, 63.61; H, 6.64. Found: C, 63.60; H, 6.45.

3β-Acetoxy-17,23-oxido-23,23-diphenylnor-5-cholene (IX).—To a solution of 3.8 g. of the dibromide (VIII) in a mixture of 50 cc. of chloroform and 100 cc. of acetic acid was added with stirring 2.0 g. of zinc dust and the mixture stirred for one hour. The chloroform was removed by vacuum distillation and the residual acetic acid solution poured into water. The crude product (3.01 g.) melting at 130–140° was recrystallized from methanol to give 1.9 g. of product melting at 163.6–164.0°; $[\alpha]^{22}D$ −94.6° (2% in $CHCl_3$).

Anal. Calcd. for $C_{37}H_{46}O_3$: C, 82.48; H, 8.61. Found: C, 82.39; H, 8.68.

3β-Hydroxy-17,23-oxido-23,23-diphenylnor-5-cholene (X).—The ether acetate (IX) was saponified by refluxing with methanolic potassium hydroxide for two hours. The product, recrystallized from methanol and dried in a vacuum oven at 110° for 20 hours, melted at 183.4–184.6°; $[\alpha]^{20}D$ −95.4° (2% in $CHCl_3$).

Anal. Calcd. for $C_{35}H_{44}O_2$: C, 84.63; H, 8.93. Found: C, 84.33; H, 9.20.

3β,17α-Dihydroxynor-5-cholenic Acid (XI).—A solution of 10 g. of the hydroxy lactone (IV) in 400 cc. of 5% ethanolic potassium hydroxide was refluxed for two hours and the mixture poured into three liters of water. Dilute hydrochloric acid was added to the solution of the potassium salt until it was just faintly acid to litmus and the precipitated free acid filtered, washed neutral, and air-dried. The crude acid (10.5 g.) was recrystallized from acetone to give 8.0 g. of needles, $[\alpha]^{21}D$ −55.3° (2% in methanol). Heating the hydroxy acid caused relactonization to take place. Thus when the melting point capillary was inserted above 250°, the acid immediately melted and bubbled up the tube, resolidified into needles on the sides of the capillary and

(17) R. Schoenheimer, *J. Biol. Chem.*, **110**, 461 (1935).

(18) Obtained from Westvaco Chlorine Products Corporation, Newark, California.

finally remelted at the melting point of the hydroxy lactone (IV) (281–282°). When the melting point capillary was inserted below 150°, only the melting point of the hydroxy lactone was obtained.

Anal. Calcd. for $C_{23}H_{36}O_4$: C, 73.36; H, 9.64. Found: C, 73.17; H, 9.71.

When titrated in aqueous methanol the hydroxy acid (XI) gave an equivalent weight of 385 instead of the expected 376.5. An examination of its infrared spectrum showed the presence of some hydroxy lactone (IV) in the analytical sample.

Methyl 3β,17α-Dihydroxynor-5-cholenate (XII).—The silver salt of the hydroxy acid (XI) was prepared by a modification of the method of Allen and Wilson.[19] To a solution of 37.6 g. of the acid (XI) and 7.15 g. of potassium hydroxide in 500 cc. of methanol was added 500 cc. of water. A solution of 18.7 g. of silver nitrate in 100 cc. of water was slowly added with agitation and the slurry of silver salt stirred for 5 minutes. Methyl iodide (142 g.) was added, the mixture stirred for 15 minutes and filtered. The product was distributed between the filter cake and the filtrate. The filter cake was extracted with hot methanol, and the extracts added to the original filtrate. The pooled liquids were concentrated to a small volume and poured into water. The product after filtration and drying below 50° (38.5 g.) was recrystallized from methanol to give 32.5 g. of fine needles with a peculiar melting point behavior. When the sample was inserted in the heating-bath at 120°, it melted at 138–140°, resolidified at 155–190° and finally remelted at 280–282°; $[\alpha]^{23}D$ −50.0° (2% in methanol). When reheated in a bath the sample melted only at 280–282° and was found to be hydroxy lactone (IV). When heated, therefore, the methyl ester apparently lost the elements of methanol to regenerate the lactone. The analytical sample was dried in a vacuum oven over phosphorus pentoxide at 55° for one hour; analysis indicated a solvate containing one-half mole of methanol.

Anal. Calcd. for $C_{24}H_{38}O_4 \cdot {}^1/_2CH_3OH$: C, 72.37; H, 9.92. Found: C, 71.69; H. 9.99.

When dried in a vacuum-oven at 100° for 16 hours, the sample lost 4.03% in weight (loss calculated for a solvate containing one-half mole of methanol, 3.95%) and had the same melting point behavior as the solvate; $[\alpha]^{20}D$ −54.2° (2% in methanol).

Anal. Calcd. for $C_{24}H_{38}O_4$: C, 73.80; H, 9.81. Found: C, 74.10; H, 10.02.

Methyl 3β-Acetoxy-17α-hydroxynor-5-cholenate (XIII).—A solution of 56.5 g. of the methyl ester (XII) in 282 cc. of dry pyridine and 57 cc. of acetic anhydride was warmed at 60° for one hour, allowed to stand overnight and then poured into water. The precipitate was filtered, washed thoroughly with water and dried at 50°, yielding 59.5 g., m.p. 174.0–181.0°. After recrystallizing from methanol, 48.7 g. of needles melting at 185.2–187.2° was obtained; $[\alpha]^{23}D$ −59.9° (2% in $CHCl_3$).

Anal. Calcd. for $C_{26}H_{40}O_5$: C, 72.19; H, 9.32. Found: C, 71.85; H, 9.43.

Methyl 3β-Acetoxynor-5,16-choladienate (XV).—A solution of 10.44 g. of the acetoxy ester (XIII) in 104 cc. of dry pyridine was cooled in an ice–salt-bath and 10.4 cc. of redistilled thionyl chloride added. The mixture was allowed to stand for two hours in the cold-bath and for one hour at room temperature, then poured into ice-water with agitation and stirred until the oily crystals hardened. The crystals were filtered and washed neutral with water. The crude product (10.0 g.) was recrystallized three times from methanol to give 4.75 g. of needles, m.p. 125.8–127.2°. The combined filtrates from the recrystallization were concentrated to a low volume, cooled and filtered. These crystals after two recrystallizations from ligroin (b.p. 90–120°) yielded needles melting at 207.2–209°. A mixture with a sample of the lactone acetate showed no depression in the melting point. It is apparent, therefore, that considerable relactonization takes place during the dehydration reaction. The analytical sample of (XV) melted at 127.8–128.6°; $[\alpha]^{22}D$ −59.4° (2 % in $CHCl_3$).

Anal. Calcd. for $C_{26}H_{38}O_4$: C, 75.32; H, 9.24. Found: C, 74.90; H, 9.33.

(19) C. F. H. Allen and C. V. Wilson, *Org. Syntheses*, **26**, 52 (1946).

3β-Hydroxynor-5,16-choladienic Acid (XVI).—A solution of 1.0 g. of the diene ester (XV) in 25 cc. of 10% ethanolic potassium hydroxide and 10 cc. of water was refluxed for 48 hours and poured into ice-water. Dilute hydrochloric acid was added until the solution was acid to congo. The precipitate was filtered and washed neutral with water. The crude product (0.84 g.), m.p. 204–212°, was recrystallized from methanol to give 0.68 g. of fine needles melting at 220.0–221.2°; $[\alpha]^{26}D$ −57.8° (2% in methanol).

Anal. Calcd. for $C_{23}H_{34}O_3$: C, 77.05; H, 9.56. Found: C, 77.00; H, 9.90.

Methyl 3β-Hydroxynor-5,16-choladienate (XVII).—This ester was prepared from the silver salt of acid (XVI) by the method given for the preparation of the ester (XII) except that it was necessary to reflux the reactants for several minutes before silver iodide precipitated. The product was recrystallized from methanol and dried under vacuum at 58° for several hours to give fine needles melting at 110.5–113.5°; $[\alpha]^{20}D$ −54° (2% in methanol). Analysis showed that the ester was a methanol solvate.

Anal. Calcd. for $C_{24}H_{36}O_3 \cdot {}^1/_2CH_3OH$: C, 75.73; H, 9.86. Found: C, 75.99; H, 9.74.

When dried in a vacuum oven for 16 hours the unsolvated ester was obtained, m.p. 105–110°.

Anal. Calcd. for $C_{24}H_{36}O_3$: C, 77.38; H, 9.74. Found: C, 77.22; H, 9.99.

Acetylation of the methyl ester using pyridine and acetic anhydride gave a crude product melting at 116–122°. Two recrystallizations from methanol gave fine needles melting at 125.6–127.0°; $[\alpha]^{25}D$ −58.9° (2% in $CHCl_3$). A mixture with a sample of methyl 3β-acetoxynor-5,16-choladienate (XV) showed no depression in the melting point.

Methyl 3β-Acetoxynor-5-cholenate (XVIII).—A solution of 3.0 g. of the diene ester (XV) dissolved in 300 cc. of absolute alcohol was hydrogenated at atmospheric pressure and room temperature using 0.6 g. of 10% palladium-on-charcoal. The reduction was stopped when 194 cc. of hydrogen had been consumed (24 minutes). The catalyst was removed by filtration and the volume reduced to 50 cc. The solution was poured into water, filtered and dried to give 2.79 g., m.p. 121–129°. The crude product was recrystallized once from acetone, once from ethanol and twice from methanol to give 1.70 g. of long needles, m.p. 132.8–133.4°; $[\alpha]^{23}D$ −40.7° (2% in $CHCl_3$). This product corresponded in its properties to the methyl 3β-acetoxynor-5-cholenate described by Plattner and Pataki.[7] A mixture with a sample of authentic ester (XVIII) prepared in this Laboratory from 3-β-acetoxybisnor-5-cholenic acid by Arndt–Eistert homologation showed no depression in the melting point. The infrared spectrum of the ester prepared by the reduction of the diene was identical with that of the ester synthesized from the bisnor acid.

Anal. Calcd. for $C_{26}H_{40}O_4$: C, 74.96; H, 9.68. Found: C, 74.70; H, 9.80.

Methyl 3β-Acetoxynor-5-cholenate (XVIII).—3β-Acetoxy-23-diazonor-5-cholene-22-one was prepared by the procedure of Wettstein.[20] The crude diazo compound (16 g.) melting at 234–235° was dissolved in methylene chloride, concentrated to a small volume and the methylene chloride completely replaced with ethyl ether by distillation. The ethyl ether solution was concentrated to a thin slurry of crystals, cooled to 5° and filtered to give 9.7 g. of purified diazo ketone, m.p. 260–265°. To a refluxing solution of 9.7 g. of the diazo ketone in 750 cc. of anhydrous methanol was added a slurry of 1.7 g. of freshly precipitated silver oxide over a two-hour period. Five grams of activated carbon was added, the mixture refluxed for one additional hour and filtered through a mat of diatomaceous earth. The filtrate was concentrated to 30 cc., cooled to 10° and filtered to give 8.1 g. of product melting at 125–129°. The crude product after several recrystallizations from methanol and ethanol gave 4.0 g. of needles, m.p. 131.8–133.4°; $[\alpha]^{22}D$ −46.7° (2% in $CHCl_3$).

3β-Hydroxynor-5-cholenic Acid (XIX).—The ester (XVIII) was saponified by refluxing with 2% ethanolic potassium hydroxide. After recrystallization from methanol, the product melted at 243.6–245.2°; $[\alpha]^{21}D$ −31.9° (2% in dioxane). The constants agree with those reported by Plattner and Pataki.[7]

(20) A. Wettstein, *Helv. Chim. Acta*, **24**, 311 (1941)

3β,23-Dihydroxy-23,23-diphenylnor-5,16-choladiene (XX).—The ester (XV) was converted to the diphenylcarbinol (XX) using the procedure given for the preparation of (VI). The crude product was recrystallized from methanol to give fine needles melting at 148–150° with bubbling; $[\alpha]^{26}D$ −50.3° (2% in $CHCl_3$). The analytical sample after drying in a vacuum-oven over phosphorus pentoxide at 55° for one hour contained methanol of solvation.

Anal. Calcd. for $C_{35}H_{44}O_2 \cdot {}^1/_2CH_3OH$: C, 83.15; H, 9.04. Found: C, 83.23; H, 8.93.

The methanol was removed by drying a sample at 100° for 18 hours at 10 μ pressure: m.p. 144.4–145.5° with bubbling.

Anal. Calcd. for $C_{35}H_{44}O_2$: C, 84.63; H, 8.93. Found: C, 84.22, 84.53; H, 9.30, 9.13.

The ethanol solvate melted at 140–143° with bubbling; $[\alpha]^{26}D$ −50.1° (2% in $CHCl_3$).

Anal. Calcd. for $C_{35}H_{44}O_2 \cdot {}^1/_2C_2H_5OH$: C, 83.19; H, 9.12. Found: C, 83.03; H, 8.92.

3β-Acetoxy-23,23-diphenylnor-5,16,22-cholatriene (XXI).—The diphenylcarbinol (XX) was dehydrated and the 3-hydroxyl acetylated by the method of Whitman and Schwenk.[21]

(21) B. Whitman and E. Schwenk, THIS JOURNAL, 68, 1865 (1946).

A mixture of 2.07 g. of the carbinol (XX), 20 cc. of acetic acid and 8.0 cc. of acetic anhydride was cooled to 10° and 0.20 cc. of perchloric acid (72%) C.P. was added with external cooling. The mixture was shaken gently until complete solution was obtained, allowed to stand at 15–20° for 30 minutes, poured into ice-water and filtered. The crude dry product, 2.05 g., m.p. 154.5–156.5°, was crystallized from methanol and then ethanol to give 1.60 g. of plates melting at 161.5–162.0°; $[\alpha]^{23}D$ −13.0° (2% in $CHCl_3$). The ultraviolet adsorption $\lambda_{max.}^{ethanol}$ 250 mμ was log ε 4.2.

Anal. Calcd. for $C_{37}H_{44}O_2$: C, 85.33; H, 8.52. Found: C, 85.42; H, 8.80.

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[CONTRIBUTION FROM THE RESEARCH LABORATORIES OF SCHIEFFELIN & CO.]

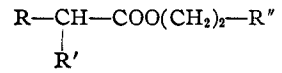
Basic Esters and Quaternary Derivatives of β-Hydroxy Acids as Antispasmodics[1]

BY GINO R. TREVES AND FRANK C. TESTA

A series of basic esters of (1-hydroxycycloalkyl)-arylacetic acids is described. Eighteen new compounds were synthesized and isolated as hydrochlorides and quaternary methiodides. Preliminary work done in our pharmacological laboratory shows that several members of our series possess neurotropic activity comparable to that of atropine and musculotropic activity of the order of magnitude of papaverine.

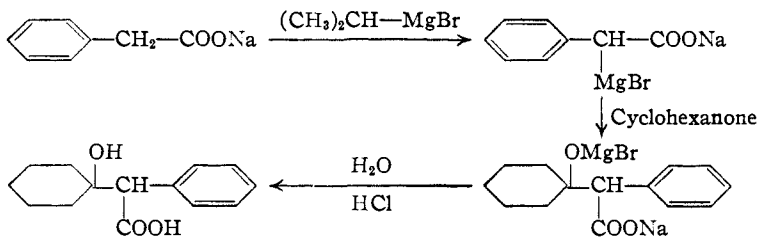
In recent years, a large number of basic esters of various hydroxy acids have been prepared and tested for pharmacological action. Among them, esters of tropic, benzilic, mandelic and substituted glycolic acids have been shown to possess high antispasmodic activity.

In a search for new and better antispasmodic agents, a series of alkylaminoalkyl and N-piperidinoalkyl esters of (1-hydroxycycloalkyl)-arylacetic acids were synthesized. These can be represented by the general structure



R is a 1-hydroxycycloalkyl group
R′ is a phenyl or a 4-methoxyphenyl group
R″ is a substituted amino group or a 1-piperidyl group

Six acids, five of which are new, were used for the syntheses of the esters. For purpose of comparison, the structural formulas of tropic acid (I), the "acid moiety" of atropine, and of (1-hydroxycyclohexyl)-phenylacetic acid (II), one of our acids, are shown above. It is observed that the carbon attached to the primary hydroxy group in I becomes part of a cyclohexyl ring in II and that the hydroxy group assumes in our structure a position angular to the ring. The acids were prepared by a modification of a method devised by Ivanoff and Spassoff.[2] The procedure exemplified for the preparation of (1-hydroxycyclohexyl)-phenylacetic acid is



Sodium phenylacetate was added to an ethereal solution of isopropylmagnesium bromide. Evolution of propane occurred with the formation of the intermediate sodium phenylacetate magnesium bromide. The addition of cyclohexanone in ether, followed by hydrolysis, produced the desired β-hydroxy acid. Table I shows the structures of the acids with their melting points, yields, and carbon hydrogen analyses.

By treating a solution of the sodium salts of the acids with the hydrochlorides of β-chloroethylalkylamines and β-chloroethyl-N-piperidine in isopropyl alcohol, eighteen new esters were synthesized and isolated as hydrochlorides. The quaternary methiodides were obtained by converting these hydrochlorides to the free bases and treating them with methyl iodide.

Tables II and III show the hydrochlorides and methiodides along with melting points, carbon–hydrogen analyses and antispasmodic activities.

(1) Presented at the Cleveland Meeting of the Division of Medicinal Chemistry, American Chemical Society, April, 1951.

(2) D. Ivanoff and A. Spassoff, *Bull. soc. chim.*, [4] 49, 375 (1931).