[CONTRIBUTION FROM THE DEPARTMENT OF CHEMISTRY, UNIVERSITY OF NOTRE DAME]

The Chemotherapy of Cancer. III.[1] The Synthesis of Dihydroxytetrahydronaphthoic Acids Related to Podophyllotoxin

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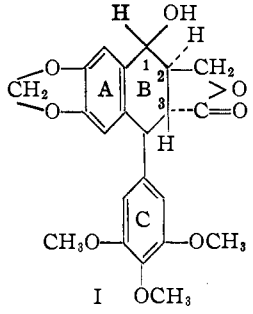
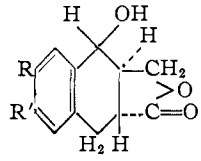
A general synthetic method is reported for the preparation of substituted *trans*-1-hydroxy-*cis*-2-methylol-*trans*-3-carboxy-1,2,3,4-tetrahydronaphthalenes, which are progenitors of lactones similar in structure to podophyllotoxin and the peltatins.

In the search for substances with selective action against neoplastic tissue, the naturally-occurring mitotic poisons are important. While these substances are, in general, too toxic to be of therapeutic value, they may serve as models for the synthesis of related and simpler compounds. Much work has been done on substances related to colchicine,[3] and recently the constituents of *podophyllum peltatum* have become of considerable interest. At least three of these components are active growth-inhibitors.[4] Recent work by Hartwell and Schrecker[5] has shown that the best-known of these substances, podophyllotoxin, probably has structure I. Hartwell and Detty[6] have shown that the other two, α-peltatin and β-peltatin, are closely related to podophyllotoxin, and have the same lactone ring system. The stereochemistry of the hydroaromatic (B) rings in these substances is intimately connected with the physiological activity, since the active compounds are readily and irreversibly epimerized at C_3 by treatment with dilute base, and the epimers have no growth-inhibitory properties. The lactone ring of the active compounds has, therefore, been assigned the *trans* configuration.[5]

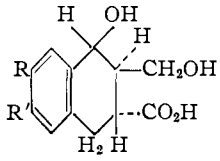


I

It seemed of interest to determine whether a compound differing from podophyllotoxin in the absence of the trimethoxyphenyl group (C ring) would retain the mitotic poisoning activity. The synthesis of lactones of type II, was, therefore, undertaken, and in this paper we are reporting the preparation of dihydroxy acids III, which are progenitors of the desired *trans* lactones.



II



III

Substituted benzylsuccinic anhydrides IV were prepared by the general procedure of Cornforth, Hughes and Lions,[7] from anisaldehyde (R = OCH_3, R′ = H), veratricaldehyde (R = R′ = OCH_3) and piperonal (R,R′ = O–CH_2–O). The succinic anhydrides were then cyclized to 3-carboxy-1-tetralones (V); in the present work it was found advantageous to use stannic chloride in place of aluminum chloride[8] to minimize cleavage of the ether groups.[9] In the case of the dimethoxy and methylenedioxybenzylsuccinic anhydrides, cyclization could occur either to the 3- or the 5-position, and could lead to a tetralone or a hydrindone. The structure of the product from dimethoxybenzylsuccinic anhydride was established by its infrared spectrum (carbonyl frequency 1664 cm.$^{-1}$), which eliminated a hydrindone structure,[10] and by oxidation to hemipic acid (VI), which was identified as its N-ethylimide; this showed that cyclization occured to the 5-position.[11]

The 3-carboxy-1-tetralones were condensed with formaldehyde to give methylol derivatives VII; it was found very difficult to control the reaction so that only one methylol group was introduced, but fair yields were obtained by using an excess of the ketone and a short reaction time. The monomethylol derivatives VII were assigned the *trans* configurations, because in every case acidification of the basic solution gave the free acid and not a lactone. If the carboxyl and methylol groups were *cis* to each other, lactonization should occur very readily, as it does in the case of podophyllic acid.[5] When excess formaldehyde was used, the monomethylol compounds were converted to dimethylol deriva-

(1) Previous paper, K. N. Campbell, A. Schrage and B. K. Campbell, *J. Org. Chem.*, **15**, 1139 (1950).

(2) Du Pont Fellow, 1949–1950. Abstracted from the Ph.D. dissertation of John A. Cella, University of Notre Dame, 1950. Part of this work was presented at the American Chemical Society meeting, Chicago, September, 1950.

(3) H. Lettre, *Angew. Chem.*, **63**, 421 (1951); J. L. Hartwell, M. V. Nadkarni and J. Leiter, THIS JOURNAL, **74**, 3180 (1952).

(4) J. Leiter, V. Downing, J. L. Hartwell and M. J. Shear, *J. Natl. Cancer Inst.*, **10**, 1273 (1950); E. M. Greenspan, J. Leiter and M. J. Shear, *ibid.*, **10**, 1295 (1950).

(5) J. L. Hartwell and A. W. Schrecker, THIS JOURNAL, **73**, 2909 (1951).

(6) J. L. Hartwell and W. E. Detty, *ibid.*, **72**, 246 (1950).

(7) J. W. Cornforth, G. K. Hughes and F. Lions, *J. Proc. Roy. Soc. N. S. Wales*, **72**, 228 (1939).

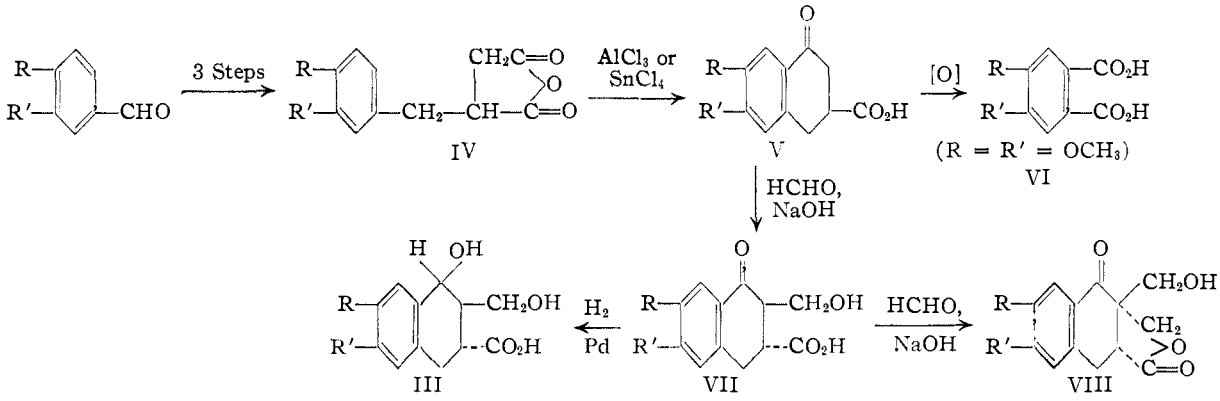
(8) R. D. Haworth, B. Jones and Y. M. Way, *J. Chem. Soc.*, 10 (1943).

(9) E. C. Horning and G. N. Walker, THIS JOURNAL, **74**, 5147 (1952), have found polyphosphoric acid a good reagent for these cyclizations.

(10) The carbonyl frequency of 5-membered cyclic ketones is much higher, 1705 cm.$^{-1}$ or above; see J. Lecomte, *J. Phys. Radium*, **6**, 257 (1945).

(11) After the present work was completed, a paper by Horning and Walker (ref. 9) reported the preparation of 6,7-dimethoxy-3-carboxyl-1-tetralone; they established the structure by dehydrogenation to the naphthalene derivative.

CHART I



tives, which could be isolated only as the lactones VIII, similar to those prepared by Haworth and Sheldrick.[12] This indicates that the second methylol group is *cis* with respect to the carboxyl group.

Reduction of the methylol keto acids VII to the carbinols III was difficult to accomplish, as frequently either poisoning of the catalyst occurred, or the reduction did not stop with the absorption of one mole of hydrogen. By use of supported palladium catalysts under carefully controlled conditions, it was possible to prepare the alcohols. In one case reduction gave two isomeric alcohols, in a ratio of about four to one. The isomer obtained in lower yield was lower-melting, and lost water at the melting point. The isomer obtained in better yield was higher melting, easily crystallized and stable to heat. The higher melting form may have the *trans* configuration (*i.e.*, the hydroxyl group on carbon atom 1 was assumed to be *trans* to the methylol group on carbon atom 2), and the lower melting isomer may have the *cis* configuration. If one considers that water is easily eliminated from substances which have a hydrogen atom *trans* to an adjacent hydroxyl group, and is eliminated with difficulty when there is no hydrogen atom *trans* to the hydroxyl group, these configurations are consistent with the properties of the isomers. Definite assignment of configuration, however, must await further work on these compounds.

Experimental[13]

Arylidenesuccinic Acids.—The aromatic aldehydes were condensed with dimethyl succinate in the presence of sodium methoxide by the general procedure of Cornforth, Hughes and Lions.[7]

Anisylidenesuccinic Acid.—From 61.2 g. of freshly distilled anisaldehyde, 65.7 g. of dimethyl succinate and 27.6 g. of sodium in 300 ml. of dry methanol there was obtained 95.7 g. of crude product, m.p. 165–175°. After two recrystallizations from ethyl acetate 85.0 g. (80%) of product, m.p. 188–191°, was obtained.

Anal. Calcd. for $C_{12}H_{12}O_5$: C, 61.01; H, 5.12. Found: C, 61.10; H, 5.50.

Veratrylidenesuccinic Acid.—The yield of product of m.p. 173–175° was 62%. Stobbe reported the melting point as 175°.[14]

Piperonylidenesuccinic Acid.—The yield of product, after recrystallization from a 4:1 mixture of ethyl acetate and ligroin (b.p. 90–120°), was 56%. The material melted at 190–194°. The literature melting point is 194–195°.[7]

Benzylsuccinic Acids.—The arylidenesuccinic acids were hydrogenated at room temperature and 100 atmospheres in slightly alkaline aqueous solution over Raney nickel, and the reduction products were isolated by acidification of the solution.

***p*-Methoxybenzylsuccinic Acid.**—The crude product, m.p. 94–97°, was obtained in 89% yield. After two recrystallizations from water (charcoal) the melting point was 100–101°, which agrees with the value reported by Haworth, Jones and Way.[8] When this acid was allowed to stand under water for several days it was converted to a polymorphic form, m.p. 113–115°; neut. equiv. calcd. 119, found 122. This material formed an anhydride identical with that obtained from the 100–101° melting form.

3,4-Dimethoxybenzylsuccinic Acid.—The product from the reduction of veratrylidenesuccinic acid formed a stable hydrate, m.p. 67–69°, which could be recrystallized from benzene.[15] The yield of pure material from 152 g. of unsaturated acid was 147 g., or 90%.

Anal. Calcd. for $C_{13}H_{18}O_7 \cdot H_2O$: C, 54.54; H, 6.34. Found: C, 54.87; H, 6.27.

3,4-Methylenedioxybenzylsuccinic Acid.—The reduction product was recrystallized from a four to one mixture of ethyl acetate and ligroin (b.p. 90–120°) to give an 85% yield of material of m.p. 135–137.5°.

Anal. Calcd. for $C_{12}H_{12}O_6$: C, 57.14; H, 4.80. Found: C, 57.03; H, 4.97.

Benzylsuccinic Anhydrides (IV).—The acids were converted to the anhydrides by the method of Haworth, Jones and Way.[8] **3,4-Dimethoxybenzylsuccinic anhydride**, m.p. 93.5–94.5°, was obtained in 87% yield.[15]

Anal. Calcd. for $C_{13}H_{14}O_5$: C, 62.39; H, 5.65. Found: C, 62.70; H, 5.72.

4-Methoxybenzylsuccinic anhydride, m.p. 90–92°, was isolated in 89% yield, and was purified by recrystallization from benzene–hexane mixture. The melting point is in agreement with the value reported by Haworth, Jones and Way.[8]

3,4-Methylenedioxybenzylsuccinic Anhydride.—When the reaction mixture was worked up immediately, and the crude product recrystallized from benzene–hexane mixture, the anhydride was obtained in 94% yield and melted at 104.5–105.5°.

Anal. Calcd. for $C_{12}H_{10}O_5$: C, 61.53; H, 4.30. Found: C, 61.48; H, 4.50.

If the reaction mixture was allowed to stand for some hours, an insoluble anhydride precipitated (yield *ca.* 94%). This product melted at 152–153.5° after recrystallization from ethyl acetate and hexane.

Anal. Calcd. for $C_{12}H_{10}O_5$: C, 61.53; H, 4.30. Found: C, 61.60; H, 4.24.

The two substances were apparently polymorphic forms of methylenedioxybenzylsuccinic anhydride, for both of

(12) R. D. Haworth and G. Sheldrick, *J. Chem. Soc.*, 289 (1941).

(13) Melting points are uncorrected. Analyses were carried out at the Micro-Tech Laboratories, Skokie, Illinois.

(14) H. Stobbe, *Ann.*, **380**, 30 (1911).

(15) Horning and Walker (ref. 9) were unable to obtain this acid as a solid; they report a melting point of 102° for the anhydride, whereas the anhydride obtained in this work melted at 93.5–94.5°. We are unable to explain these discrepancies, since both anhydrides gave dimethoxy-3-carboxy-1-tetralone melting at 226°, unless it is another case of the polymorphism so frequently encountered in these series.

them could be hydrolyzed to the original succinic acid, and the lower-melting form (104.5–105.5°) was converted to the higher-melting form by seeding its ethyl acetate solution with a few crystals of the latter.

Substituted 3-Carboxy-1-tetralones (V). 3-Carboxy-1-keto-7-methoxy-1,2,3,4-tetrahydronaphthalene (V, R = OCH_3, R′ = H).—This was prepared by a modification of the method of Haworth, Jones and Way.[8] A solution of 72 g. (0.33 mole) of 4-methoxybenzylsuccinic anhydride in 300 ml. of nitrobenzene was treated with 88 g. (0.7 mole) of anhydrous aluminum chloride at such a rate that the temperature of the reaction mixture did not rise above 35°, and the mixture was allowed to stand at room temperature for four hours. The mixture was hydrolyzed, the nitrobenzene removed by steam distillation and the residue was recrystallized twice from water (decolorizing carbon). There was obtained 29 g. (42%) of product, m.p. 149–151°. The literature melting point[5] is 151°. The methyl ester, prepared by refluxing the acid in dry methanol containing a trace of sulfuric acid, melted at 78–79.5°.

3-Carboxy-6,7-dimethoxy-1-keto-1,2,3,4-tetrahydronaphthalene (V, R = R′ = OCH_3).—Stannic chloride (219 g., 0.84 mole) was added dropwise, at room temperature, to a solution of 91 g. (0.4 mole) of 3,4-dimethoxybenzylsuccinic anhydride in 500 ml. of nitrobenzene and the mixture was allowed to stand at room temperature for 30 minutes. It was poured into 200 g. of cracked ice and 50 ml. of concentrated hydrochloric acid, and 500 ml. of ligroin (b.p. 90–120°) was added to dilute the nitrobenzene layer. The product solidified at the interface, and was collected by filtration. It was recrystallized from methyl cellosolve to give 76.3 g. (85%) of material, m.p. 224–227°.

Anal. Calcd. for $C_{13}H_{14}O_5$: C, 62.39; H, 5.64. Found: C, 62.47; H, 5.71.

A mixture of 1.0 g. of the tetralone, 4 g. of potassium permanganate, 1 ml. of 10% sodium hydroxide and 80 ml. of water was refluxed for two hours, then acidified with sulfuric acid and refluxed an additional 30 minutes. The cooled mixture was treated with sodium bisulfite to dissolve manganese dioxide, and 20 ml. of 70% ethylamine was added. The entire mixture was evaporated to dryness on a steam-bath and the residue sublimed. The sublimate was recrystallized twice from ethanol to give 400 mg. of N-ethyl-*m*-hemipimide, m.p. 228–230°. This melting point is the same as that reported by Karrer and Schmid[16] for the characteristic derivative of hemipic acid.

Anal. Calcd. for $C_{12}H_{13}O_4N$: C, 61.27; H, 5.56. Found: C, 61.13; H, 5.62.

3-Carboxy-1-keto-6,7-methylenedioxy-1,2,3,4-tetrahydronaphthalene (V, R,R′ = O–CH_2–O).—This was prepared as described for the dimethoxy analog, and the product was recrystallized from methyl cellosolve to give an 80% yield of material melting at 234.5–235.5°.

Anal. Calcd. for $C_{12}H_{10}O_5$: C, 61.53; H, 4.30. Found: C, 61.37; H, 4.51.

The semicarbazone melted at 265° (dec.) after recrystallization from methyl cellosolve.

Anal. Calcd. for $C_{13}H_{13}O_5N$: N, 14.43. Found: N, 14.72.

Condensation of 3-Carboxy-1-tetralones with Formaldehyde.—When an excess of the keto-acid was treated with alkaline formaldehyde solution at 10°, two products were usually obtained; the monomethylol product VII and the lactone of the dimethylol product VIII. The reaction mixture was worked up as follows: It was acidified to *p*H 4 with glacial acetic acid, and the unreacted starting material which precipitated at this point was removed. The filtrate was then acidified to *p*H 2 with hydrochloric acid, and about 20 ml. of ethyl acetate was added to induce crystallization. The precipitate obtained at this point was the monomethylol derivative, which was collected and recrystallized from 95% ethanol. The lactone of the dimethylol derivative was isolated by extracting the filtrate with ethyl acetate. The extract was washed with dilute sodium bicarbonate solution and then with water, dried over magnesium sulfate and evaporated to dryness. The residue was recrystallized from water.

3-Carboxy-1-keto-7-methoxy-1,2,3,4-tetrahydronaphthalene and Formaldehyde.—A mixture of 5.5 g. (0.025 mole) of the 3-carboxy-1-tetralone, 1.9 g. (0.0025 mole) of 37% formalin, 5 g. of sodium hydroxide and 125 ml. of water was kept at 10° for one hour, and worked up as described above. There was obtained 1.3 g. (20.8%) of the monomethylol derivative, *trans*-3-carboxy-2-hydroxymethyl-1-keto-7-methoxy-1,2,3,4-tetrahydronaphthalene (VII, R = OCH_3, R′ = H), m.p. 153–155°.

Anal. Calcd. for $C_{13}H_{14}O_5$: C, 62.47; H, 5.83. Found: C, 62.40; H, 5.64.

There was also obtained 0.25 g. of a neutral compound, the lactone of the dimethylol derivative VIII, m.p. 137–138.5°.

Anal. Calcd. for $C_{14}H_{14}O_5$: C, 64.11; H, 5.39. Found: C, 64.26; H, 5.38.

3-Carboxy-6,7-dimethoxy-1-keto-1,2,3,4-tetrahydronaphthalene and Formaldehyde.—From 18 g. (0.072 mole) of the 3-carboxy-1-tetralone, 1.2 g. (0.004 mole) of paraformaldehyde and 3.2 g. (0.08 mole) of sodium hydroxide in 100 ml. of water, kept at 10° for ten hours, there was recovered 10 g. of starting material, 4.3 g. of the monomethylol derivative (VII, R = R′ = OCH_3) and 0.25 g. of the lactone of the dimethyl derivative.

trans-3-Carboxy-6,7-dimethoxy-2-hydroxymethyl-1-keto-1,2,3,4-tetrahydronaphthalene (VII, R = R′ = OCH_3) was isolated as the monohydrate, m.p. 125°.

Anal. Calcd. for $C_{14}H_{16}O_6 \cdot H_2O$: C, 56.30; H, 6.05. Found: C, 55.83; H, 5.90.

It could be converted to the anhydrous form, m.p. 185°, by careful drying *in vacuo*.

Anal. Calcd. for $C_{14}H_{16}O_6$: C, 60.00; H, 5.75. Found: C, 60.00; H, 5.70.

3-Carboxy-6,7-dimethoxy-2,2-dimethylol-1-keto-1,2,3,4-tetrahydronaphthalene *cis* lactone (VIII, R = R′ = OCH_3), melted at 170–171°.

Anal. Calcd. for $C_{15}H_{16}O_6$: C, 61.63; H, 5.52. Found: C, 61.75; H, 5.60.

3-Carboxy-1-keto-6,7-methylenedioxy-1,2,3,4-tetrahydronaphthalene and Formaldehyde.—From 9.4 g. (0.04 mole) of the carboxytetralone, 1.8 g. (0.03 mole) of 37% formalin, 3.2 g. (0.08 mole) of sodium hydroxide and 100 ml. of water, kept at 10° for seven hours, there was obtained 5 g. of unreacted starting material, 1.3 g. (26%) of the monomethylol compound and 0.5 g. of the dimethylol lactone.

trans-3-Carboxy-2-hydroxymethyl-1-keto-6,7-methylenedioxy-1,2,3,4-tetrahydronaphthalene (VII, R,R′ = O–CH_2–O–) was obtained as a white, crystalline solid, m.p. 183.5–185°.

Anal. Calcd. for $C_{13}H_{12}O_6$: C, 59.09; H, 4.58. Found: C, 59.32; H, 4.68.

3-Carboxy-2,2-dimethylol-1-keto-6,7-methylenedioxy-1,-2,3,4-tetrahydronaphthalene *cis* lactone (VIII, R,R′ = O–CH_2–O–) was obtained as a white solid, m.p. 173–174°.

Anal. Calcd. for $C_{14}H_{12}O_6$: C, 60.87; H, 4.38. Found: C, 61.30; H, 4.48.

Hydrogenation of 3-Carboxy-2-hydroxymethyl-1-keto-7-methoxy-1,2,3,4-tetrahydronaphthalene.—When 5.0 g. of this compound in 75 ml. of ethanol was shaken with hydrogen at 4 atmospheres and 60° in the presence of 50 mg. of 10% palladium on charcoal, one molar equivalent of hydrogen was absorbed in 90 minutes. The catalyst was removed by filtration of the hot solution, and the filtrate was concentrated under reduced pressure to 25 ml. On cooling the solution there precipitated 3.8 g. (75.5%) of the carbinol (III, R = OCH_3), m.p. 163–165°.

Anal. Calcd. for $C_{13}H_{16}O_5$: C, 61.89; H, 6.39. Found: C, 61.80; H, 6.62.

A small amount of an isomeric carbinol, m.p. 150–152° (dec.) was obtained by evaporating the filtrate to dryness and recrystallizing the residue from water.

Anal. Calcd. for $C_{13}H_{16}O_5$: C, 61.89; H, 6.39. Found: C, 61.90; H, 6.40.

Hydrogenation of 3-carboxy-6,7-dimethoxy-2-hydroxymethyl-1-keto-1,2,3,4-tetrahydronaphthalene (600 mg.) was accomplished by treating it in 50 ml. of ethanol with hydrogen at 4 atmospheres and 50° in the presence of 100 mg. of 5% palladium on alumina. One equivalent of hydrogen was absorbed in 90 minutes. The filtrate from removal of the catalyst was evaporated to dryness and the residual oil

(16) H. Schmid and P. Karrer, *Helv. Chim. Acta*, **28**, 722 (1945)

was taken up in absolute alcohol and treated with anhydrous ether. There was obtained 520 mg. (84%) of the carbinol (III, R = R′ = OCH_3), m.p. 147° (dec.).

Anal. Calcd. for $C_{14}H_{18}O_6$: C, 59.56; H, 6.43. Found: C, 59.53; H, 6.57.

3-Carboxy-2-hydroxymethyl-1-keto-6,7-methylenedioxy-1,2,3,4-tetrahydronaphthalene (1.0 g.) in 75 ml. of ethanol was shaken with hydrogen at 60° and 4 atmospheres in the presence of 200 mg. of 5% palladium on alumina for 60 minutes, at the end of which time one equivalent of hydrogen had been absorbed and the reaction had stopped. The oil remaining after removal of the catalyst and evaporation of the solvent was crystallized from 10% aqueous ethanol to give 0.8 g. (80%) of the carbinol (III, R,R′ = O-CH_2O-), m.p. 157° (dec.).

Anal. Calcd. for $C_{13}H_{14}O_6$: C, 58.64; H, 5.30. Found: C, 58.76; H, 5.47.

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NOTRE DAME, INDIANA

[CONTRIBUTION FROM THE RESEARCH LABORATORIES OF CHAS. PFIZER AND CO., INC.]

Magnamycin. A New Antibiotic[1,2]

BY RICHARD L. WAGNER, F. A. HOCHSTEIN, KOTARO MURAI, N. MESSINA AND PETER P. REGNA

RECEIVED MAY 25, 1953

The new antibiotic Magnamycin has been isolated from fermentation broths by solvent extraction and purified by repeated crystallization. The high purity of the weak base obtained by this procedure has been demonstrated by solubility analysis, by countercurrent distribution studies and by paper chromatography. The chemical and physical properties of Magnamycin, of its carbonyl derivatives and its tetrahydro derivative suggest the presence of both saturated and α,β-unsaturated carbonyl systems. Alkaline degradation of the antibiotic has yielded acetic acid, isovaleric acid and dimethylamine. Mild acid hydrolysis cleaves Magnamycin into two fragments, namely, the isovalerate ester of a $C_7H_{14}O_4$ compound, and a crystalline solid, the formula of which is probably $C_{29-30}H_{47-49}NO_{12}$.

Magnamycin[3] is a new antibiotic elaborated by strains of the microörganism *Streptomyces halstedii*. The antibiotic may be isolated from filtered fermentation broth by simple extraction with a water-immiscible solvent. The crude product obtained on concentration of the solvent extracts can be purified by crystallization from alcohol–water mixtures. The purity of Magnamycin obtained by repeated crystallizations from ethanol has been shown by solubility analysis to be 99.2 ± 0.1%. Both countercurrent distribution and paper chromatography studies confirm the homogeneity of this material.

On the basis of analyses of Magnamycin and its crystalline derivatives, the antibiotic has been assigned a tentative empirical formula $C_{41-42}H_{67-69}NO_{16}$. The optically active substance $[\alpha]^{25}D$ −58.6° (*c* 1%, chloroform) crystallizes from ethanol as colorless laths. It is readily soluble in most organic solvents but virtually insoluble in hexane and in water. Although Magnamycin is a weak base, pK_b 7.2, it forms stable salts with mineral acids. The water-soluble hydrochloride as well as the relatively insoluble periodate have been prepared.

Crystalline Magnamycin shows no signs of decomposition after storing for several months in the dark at room temperature. Although aqueous solutions of the new antibiotic between *p*H 5–7 show no loss in microbiological activity after 11 days at 25°, more acid solutions, *p*H 3, and more basic solutions, *p*H 9, are half-inactivated in the same period.

Although Magnamycin contains nitrogen, it gives negative ninhydrin and Van Slyke nitrogen tests, indicating the absence of any primary amine. Fehling and Tollens tests are positive. The presence of carbonyl groupings is suggested by the positive 2,4-dinitrophenylhydrazine test. The unsaturated character of Magnamycin is demonstrated further by the positive Baeyer permanganate test and the ready decolorization of bromine. The ceric nitrate test shows the presence of hydroxyl groups, while the negative ferric chloride test shows them to be non-phenolic. The boric acid test suggests that there are no suitable oriented adjacent hydroxyl groups. In strong mineral acids, *e.g.*, 2–6 *N*, Magnamycin develops a characteristic deep violet color which slowly fades to a red-brown. This characteristic reaction in aqueous acid interferes with many standard diagnostic color tests.

Several biologically active derivatives of Magnamycin have been prepared. A diacetyl derivative is formed on acetylation in acetic anhydride–pyridine. An oxime and a thiosemicarbazone have been prepared by standard procedures. The hydrogenation of Magnamycin over palladium–charcoal catalyst in ethanol solution results in the prompt absorption of two moles of hydrogen and yields a crystalline tetrahydro derivative which also forms a diacetate on acetylation.

Alkaline hydrolysis of Magnamycin yields one mole each of acetic and isovaleric acids and dimethylamine. Controlled mild acid hydrolysis results in hydrolytic cleavage of the Magnamycin molecule into two fragments. The first product, a viscous oil, with the empirical formula $C_{12}H_{22}O_5$, contains the isovaleric acid of Magnamycin. The second fragment, a crystalline solid, is a weak base which appears to have the empirical formula $C_{29-30}H_{47-49}NO_{12}$. Although analyses of the compound itself have not been entirely satisfactory, the diacetate is readily purified, and its analyses are consistent with those calculated for the C_{29}

(1) Magnamycin is a Chas. Pfizer and Co. trademark for the antibiotic carbomycin.

(2) Presented before the Division of Medicinal Chemistry at the 123rd Meeting of the American Chemical Society, Los Angeles, Calif., March, 1953.

(3) F. W. Tanner, A. R. English, T. M. Lees and J. B. Routien, *Antibiotics and Chemotherapy*, 2, 441 (1952).