Carbobenzoxy-D- and L-serylglycyl-L-histidine Methyl Ester (III).—This product was prepared by modification of the general method of Brand and associates[13] for carbobenzoxytripeptide esters. The intermediate azide was water soluble, but was extracted with chloroform. The combined dried extracts were then added to a solution of 1.9 g. (0.011 mole) of L-histidine methyl ester in 50 ml. of chloroform. A yellow oil began to separate in a few minutes, and the mixture was allowed to stand overnight. The chloroform was decanted and replaced by dry ether. Rubbing under this solvent gave a pale yellow powder; yield 3.4 g. (96%). The product was quite hygroscopic and very soluble in water and in alcohols. For analysis it was dissolved in dry methanol and poured with stirring into 10–15 volumes of dry ether, and then filtered quickly and stored over phosphorus pentoxide. It was dried for several hours at 36° *in vacuo* over phosphorus pentoxide immediately prior to analysis; m.p. 37–55°.

Anal. Calcd. for $C_{20}H_{25}N_5O_7$: C, 53.68; H, 5.62; N, 15.69. Found: C, 53.3; H, 5.7; N, 16.4.

Insoluble precipitates were obtained with mercuric chloride and silver nitrate; the mercuric chloride complex was formed by addition of excess mercuric chloride to an aqueous solution of the peptide derivative. The precipitate was washed successively with water, methanol and ether and then dried *in vacuo;* it slowly decomposed in the range 70–160°.

Anal. Calcd. for $C_{20}H_{25}N_5O_7 \cdot 2HgCl_2$: N, 7.07; Cl, 14.32. Found: N, 7.1; Cl, 14.2.

Phthaloylhistidine was prepared in accordance with the directions of Keil.[5] The product, obtained repeatedly in 80% yield, melted at 294–296° rather than 188° as reported.[5] For analysis it was recrystallized from water.

Anal. Calcd. for $C_{14}H_{11}N_3O_4$: C, 58.94; H, 3.89; N, 14.73. Found: C, 58.9; H, 3.8; N, 14.5.

Phthaloylhistidine methyl ester hydrochloride was prepared following the directions of Keil.[5] The hydrochloride melted at 221–223° in a capillary, or 209–211° on an aluminum block. Keil[5] gives the melting point as 238–240° (uncor.). For analysis the product was recrystallized from methanol.

Anal. Calcd. for $C_{15}H_{13}N_3O_4 \cdot HCl$: C, 53.66; H, 4.20; N, 12.51. Found: C, 53.2; H, 4.1; N, 12.2.

Phthaloylhistidine methyl ester, liberated by means of potassium carbonate, melted at 187°; Keil reports 187° (uncor.).[5]

Phthaloylglycine azide was prepared from phthaloylglycyl chloride[14] and sodium azide in aqueous acetone by application of the general method described by Smith.[15] Yields of crude product, m.p. 97–99° dec., were essentially quantitative. The compound was thermally unstable, but some purification was effected by precipitating it from benzene solution by the addition of petroleum ether.

Anal. Calcd. for $C_{10}H_6N_4O_3$: C, 52.18; H, 2.62; N, 24.34. Found: C, 53.2; H, 2.3; N, 23.6.

Phthaloylglycyl-L-serine methyl ester was prepared in 68% yield from equimolar portions of the azide and L-serine methyl ester in tetrahydrofuran; m.p. after crystallization from methanol, 221–222°.

Anal. Calcd. for $C_{14}H_{14}N_2O_6$: C, 54.90; H, 4.61; N, 9.15. Found: C, 54.9; H, 4.6; N, 8.9.

Three hours of refluxing with one equivalent of 1 *N* methanolic hydrazine followed by filtration of the phthaloylhydrazide and evaporation of the solution gave quantitative yields of a crystalline residue, melting at 215–217° after crystallization from methanol. Abderhalden and Bahn[16] report 218–220° as the melting point of the DL-serylglycine anhydride.

Anal. Calcd. for $C_5H_8N_2O_3$: C, 41.66; H, 5.60; N, 19.44. Found: C, 42.1; H, 5.6; N, 19.2.

Phthaloylglycyl-L-histidine methyl ester was prepared in 76% yield from equimolar portions of phthaloylglycine azide and L-histidine methyl ester in chloroform. After crystallization from methanol it melted at 199–201°.

Anal. Calcd. for $C_{17}H_{16}N_4O_5$: C, 57.30; H, 4.53; N, 15.72; O, 23.45. Found: C, 57.0; H, 4.4; N, 15.0; O (direct), 23.4.

Phthaloylglycyl-L-histidine was prepared in 45% yields from the azide and L-histidine in 50% aqueous dioxane following the general method of Kroll.[17] The product melted at 258° dec. after crystallization from water; Turner[18] reports 258–262°.

***o*-Nitrophenoxyacetyl-L-histidine hydrazide** was prepared by minor modification of the general directions of Holley and Holley[6] from chloroform solutions of 4.8 g. (0.022 mole) of *o*-nitrophenoxyacetyl chloride and 3.7 g. of L-histidine methyl ester (0.022 mole) with 2 g. (0.022 mole) of 2,6-lutidine as the acid acceptor. After filtration of the lutidine hydrochloride the solution was evaporated to dryness. The residue was taken up in 50 ml. of methanol, treated with 1 g. of 95% hydrazine and allowed to stand overnight. There was obtained 3 g. of colorless crystals, m.p. 193–195° (39% yield). Recrystallization from 90% methanol raised the melting point to 201–202° dec.

Anal. Calcd. for $C_{14}H_{16}N_6O_5$: C, 48.27; H, 4.63; N, 24.13. Found: C, 48.7; H, 4.6; N, 22.7.

(13) E. Brand, *et al.*, THIS JOURNAL, **73**, 3510 (1951).

(14) J. C. Sheehan and V. S. Frank, *ibid.*, **71**, 1856 (1949).

(15) P. A. S. Smith, "Organic Reactions," Vol. 3, John Wiley and Sons, Inc., New York, N. Y., 1946, p. 387.

(16) E. Abderhalden and A. Bahn, *Z. physiol. Chem.*, **234**, 181 (1935).

(17) H. Kroll, Abs. Meeting Am. Chem. Soc., Sept., 1952, p. 44C.

(18) R. A. Turner, THIS JOURNAL, **75**, 2388 (1953).

EMERYVILLE, CALIFORNIA

[CONTRIBUTION FROM THE RESEARCH LABORATORIES OF CHAS. PFIZER & COMPANY, INC.]

Magnamycin B, a Second Antibiotic from *Streptomyces halstedii*

BY F. A. HOCHSTEIN AND KOTARO MURAI

RECEIVED APRIL 29, 1954

Magnamycin B is a new antibiotic which has been isolated, along with Magnamycin, from fermentation beers of *Streptomyces halstedii*. The new antibiotic resembles Magnamycin in its antibacterial spectrum, its low toxicity, and in its fundamental chemical structure. Spectral studies show it to contain an $\alpha,\beta,\gamma,\delta$-unsaturated carbonyl system, in contrast to the α,β-unsaturated carbonyl group of Magnamycin. Several microbiologically active derivatives of Magnamycin B have been prepared.

Magnamycin,[1] an elaboration product of a strain of *Streptomyces halstedii*, has been carefully characterized, and is described in earlier publications.[2–4] We wish to report at this time a second antibiotic Magnamycin B, which is elaborated by this actinomycete.

Solubility analyses and countercurrent distribution studies on samples of crude Magnamycin revealed the presence of small amounts of other antibacterial substances, substances which were more soluble in benzene and alcohols. Examination of mother liquors resulting from the crystallization

(1) Magnamycin is the trade-mark of Chas. Pfizer & Co., Inc., for the antibiotic carbomycin.

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

(3) R. L. Wagner, F. A. Hochstein, K. Murai, N. Messina and P. P. Regna, THIS JOURNAL, **75**, 4684 (1953).

(4) J. D. Dutcher, J. Vandeputte, S. Fox and L. Heuser, *Antibiotics and Chemotherapy*, **3**, 910 (1953).

of crude Magnamycin led to the isolation of Magnamycin B as a crystalline compound.

The new antibiotic crystallizes from acetone as colorless anisotropic plates, which are frequently hexagonal in outline. It melts with decomposition at 141–144°, and has $[\alpha]^{25}D$ −35°. The ultraviolet absorption spectrum (Fig. 1) shows a strong absorption peak at 278 mμ, and the infrared absorption spectrum (Fig. 2) is characteristic. Analyses of Magnamycin B and of some of its derivatives suggest a molecular formula $C_{41-2}H_{67-9}NO_{15}$. We cannot exclude completely formulas containing an additional atom of oxygen. It follows that the new antibiotic may be isomeric with Magnamycin.

The antibacterial spectrum of Magnamycin B closely resembles that of Magnamycin. A comparison of the two compounds is given in Table I.

TABLE I

MINIMUM INHIBITORY CONCENTRATIONS OF MAGNAMYCIN AND MAGNAMYCIN B

Organism	μg./per ml. Magnamycin B	Magnamycin
Escherichia coli	100	100
Psuedomonas aeruginosa	>100	>100
Proteus, sp.	>100	100
Bacillus subtilis	0.39	0.39
Streptococcus faecalis	.19–0.39	.39–0.78
Micrococcus pyogenes, var. aureus	.78	.78
Mycobacterium #607	.39	.78

The LD_{50} for mice receiving Magnamycin B hydrochloride intravenously is 300 mg./kg. body weight. Magnamycin B is therefore somewhat more toxic than Magnamycin when administered by this route.[2] The acute toxicities of the two antibiotics by intraperitoneal, intramuscular, subcutaneous and oral routes are, however, strictly comparable.

Preliminary chemical studies on the new antibiotic also show it to be closely related to Magnamycin. Both compounds are weak bases, both contain a single methoxyl group, a dimethylamino nitrogen group, and a volatile acid other than isovaleric. Both compounds yield methylmycarose isovalerate[3,5] on hydrolysis in methanolic hydrochloric acid. Vigorous aqueous acid hydrolysis of either antibiotic yields a new desoxydimethylamino sugar, $C_8H_{17}NO_4$, which we have named mycaminose. Catalytic hydrogenation of Magnamycin B shows the presence of two carbon–carbon double bonds.

Spectrophotometric studies, however, have revealed a point of structural difference in the two compounds. The ultraviolet absorption spectrum of Magnamycin B, which shows a single peak at 278 mμ, ε about 25,000, is characteristic of an α,β,γ,δ-unsaturated ketone.[6] The occurrence of peaks at 5.98, 6.16, 6.29, 10.03 and 10.25 μ in the infrared absorption spectrum is in complete accord with this structural assignment. Magnamycin, on the other hand, appears to contain an α,β-unsaturated ketone system, though it too shows the presence of two carbon–carbon double bonds on hydrogenation.[3]

(5) P. P. Regna, F. A. Hochstein, R. L. Wagner and R. B. Woodward, THIS JOURNAL, **75**, 4625 (1953).

(6) L. K. Evans and A. E. Gillam, *J. Chem. Soc.*, 432 (1953).

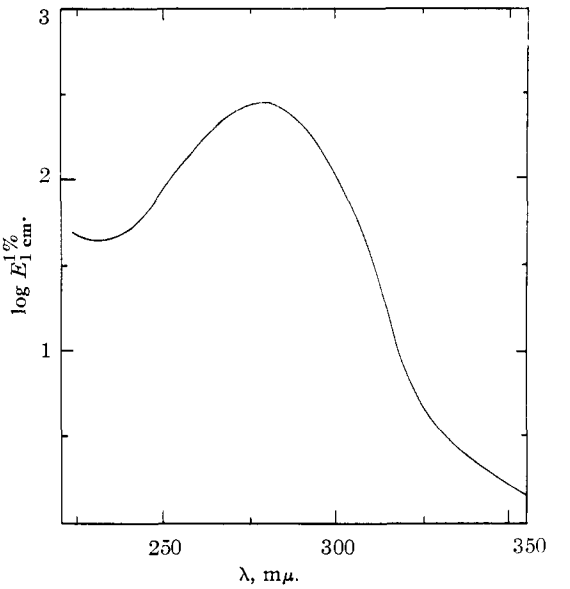


Fig. 1.—Ultraviolet absorption spectrum of Magnamycin B in absolute ethanol.

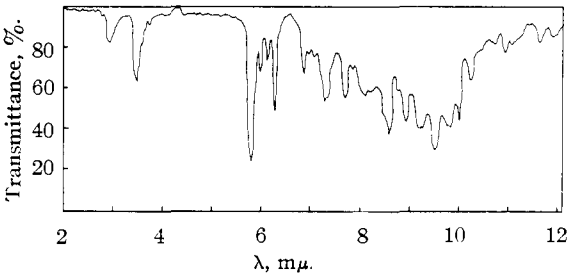


Fig. 2.—Infrared absorption spectrum of Magnamycin B in chloroform solution.

Magnamycin B has yielded a number of derivatives with biological activity of the same order of magnitude as the base compound. These include a monoacetyl derivative, a tetrahydro derivative, and a monoacetyltetrahydro derivative.

It is difficult, in view of the dearth of definitive information regarding the empirical formulas of the two antibiotics to draw precise conclusions regarding their structural relationship. Since tetrahydromagnamycin and tetrahydromagnamycin B are not identical, it seems likely that the parent compounds differ in more than double bond position. The fact that Magnamycin B and tetrahydromagnamycin B yield monoacetyl derivatives under conditions which yield diacetyl derivatives of Magnamycin and tetrahydromagnamycin confirms this opinion, and suggests further that Magnamycin B may contain one atom of oxygen less than does Magnamycin.

Experimental

Melting points were determined in capillary tubes, and are corrected. Calculated analyses have been given for a base formula $C_{41}H_{67}NO_{15}$. Equally satisfactory analyses can be calculated for certain closely related formulas, *e.g.*, $C_{41}H_{69}NO_{15}$. Optical rotations were determined in 1% chloroform solution, unless otherwise specified.

Magnamycin B.—A large experimental batch of crude Magnamycin, isolated by the solvent extraction procedure described in reference 3, was found by solubility analysis

to be only 90% pure. Bioassays suggested that the impurity was microbiologically active. The methanol–water mother liquors from a recrystallization of this product were shown by solubility analysis and countercurrent distribution to contain the major portion of the impurities. These mother liquors were diluted with a large volume of water, the amorphous precipitate dried, and used as source material for the new compound. Fifty grams of this material was dissolved in 100 ml. of anhydrous ethanol, and the viscous solution was stirred at room temperature for 8 hours. Seven grams of crude crystalline Magnamycin, which precipitated during this period, was separated by filtration. The filtrate was concentrated to substantial dryness *in vacuo*, on a steam-bath. The semi-solid residue was dissolved in 45 ml. of acetone, and the acetone solution diluted with water (50 ml.) until turbid. Crude crystalline Magnamycin B (18 g.) separated overnight. This material was further purified by three crystallizations from 5:1 acetone–water, and a final recrystallization from anhydrous acetone. The final crystallization effected no further purification, as measured by the intensity of the 278 mμ ultraviolet absorption peak and by the constancy of optical rotation. This product appeared homogeneous when examined on several paper chromatographic systems. A sixty-plate countercurrent distribution between benzene–*p*H 4.5 acetate buffer permitted ready resolution of Magnamycin B (K = 12) from Magnamycin K = 7), and showed no evidence of impurities in our analytical sample. Solubility analysis in benzene at 36.6° showed a solubility of 132.5 mg. per gram of solvent, and indicated the analytical sample to be better than 98% pure. This product was dried to constant weight at 80°, 0.1 mm. for analysis; m.p., softens at 138°, melts at 141–144° with decomposition, $[\alpha]^{25}D$ −35°.

Anal. Calcd. for $C_{41}H_{67}NO_{15}$: C, 60.50; H, 8.30; N, 1.72; methoxyl (one), 3.81; N-CH_3 (2), 3.69; mol. wt., 814. Found: C, 60.55; H, 8.42; N, 1.78; methoxyl, 4.01; N-CH_3, 3.10; volatile acids, 2.05 moles/mole.

Titration in 5:1 dimethylformamide–water shows an equivalent weight of 870, pK_b 7.56. The ultraviolet absorption spectrum in absolute ethanol has λ_{max} 278 mμ, $E^{1\%}_{1cm}$ 276, and λ_{min} 230 mμ, $E^{1\%}_{1cm}$ 35 (Fig. 1). The infrared absorption spectrum in 2% chloroform solution, 0.1-mm. cells, is shown in Fig. 2. Magnamycin B has a solubility in ethanol of about 450 mg./ml. at 25°; water dissolves 0.1–0.2 mg./ml. at this temperature. Aqueous solutions buffered at *p*H 5 have a half-life of greater than 3 months at room temperature. Strongly acid (*p*H 3) and alkaline solutions (*p*H 10) are less stable and have half-lives of about 10 days.

Magnamycin B hydrochloride was prepared by dissolving 0.42 g. of Magnamycin B in 5.5 ml. of 0.10 N hydrochloric acid at 25°. The solution was seeded, and precipitated 0.10 g. of Magnamycin B hydrochloride as very fine needles. The product was dried at 0.3 mm., 25° for 18 hours for analysis; m.p. 164–166 dec. when placed in bath at 150°, temperature rising 3° per minute.

Anal. Calcd. for $C_{41}H_{67}NO_{15}$·HCl: C, 57.92; H, 8.06, N, 1.65; Cl, 4.17. Found: C, 57.61; H, 7.84; N, 1.77; Cl, 4.15. The infrared spectrum in chloroform solution resembles that of the free base closely. Minor differences were apparent at 3.7 μ, and in the 8.5–10 μ region.

Acetylmagnamycin B was prepared by dissolving 2 g. of Magnamycin B in 2 ml. of pyridine and 2 ml. of acetic anhydride. After standing 72 hours at 26°, the volatile reactants were removed *in vacuo* at 40–50°, the viscous residue suspended in 50 ml. of water, and adjusted to *p*H 8.5. The insoluble product was separated by filtration and crystallized once from ethanol, twice from acetone–water to yield 0.5 g. of pure needles, m.p. 149–150°. After drying at 80°, 0.1 mm., this biologically active product melted at 195–205°, with much prior darkening and decomposition, $[\alpha]^{25}D$ −67°.

Anal. Calcd. for $C_{43}H_{69}NO_{16}$: C, 60.33; H, 8.13; N, 1.64. Found: C, 60.27; H, 7.92; N, 1.63; volatile acids, 3.00 moles. The ultraviolet absorption spectrum in ethanol solution shows a single peak λ_{max} 278 mμ, $E^{1\%}_{1cm}$ 240.

Tetrahydromagnamycin B.—Magnamycin B, 2.10 g., in 60 ml. of anhydrous ethanol was hydrogenated over 0.5 g. of 5% palladium–charcoal catalyst. Two equivalents (110 ml. at S.T.P.) of hydrogen was absorbed within one hour, and absorption then stopped. The amorphous product obtained on removing the ethanol *in vacuo* was twice recrystallized, with difficulty, from 1:4 toluene–hexane to yield 220 mg. of colorless product, m.p. 99–101°. This product was not analyzed.

Tetrahydromagnamycin B Hydrochloride.—Amorphous tetrahydromagnamycin B dissolves readily in 0.1 N hydrochloric acid, and precipitates the crystalline hydrochloride as short prisms, which have a solubility in water of about 35 mg./ml. at 25°, $[\alpha]^{25}D$ −42°. The ultraviolet spectrum shows a weak peak at 260–270 mμ. The infrared spectrum shows a single broad unresolved peak in the carbonyl region at 5.8 μ.

A sample[7] was recrystallized from acetone and dried *in vacuo* at 56° for 4 hours for analysis; m.p. 143–144°.

Anal. Calcd. for $C_{41}H_{71}NO_{15}$·HCl: C, 57.63; H, 8.49; N, 1.64; Cl, 4.15. Found: C, 57.82; H, 8.58; N, 1.59; Cl, 4.18.

Acetyltetrahydromagnamycin B.—Amorphous tetrahydromagnamycin B 1.5 g. was acetylated by the procedure described for acetylmagnamycin B. The product was crystallized twice from ethanol–water, once from cyclohexane, and again from ethanol–water to yield 100 mg. of purified product, m.p. 109–110.5°. This biologically active product was dried at 80°, 0.1 mm., for analysis; $[\alpha]^{25}D$ −73°.

Anal. Calcd. for $C_{43}H_{73}NO_{16}$: C, 60.05; H, 8.55; N, 1.63. Found: C, 60.48; H, 8.68; N, 1.65; volatile acids, 3.16 moles/mole.

Acid Hydrolysis of Magnamycin B.—A solution of 5.0 g. Magnamycin B in 25 ml. of methanol and 2.5 ml. of concentrated hydrochloric acid was held at 5° for 17 hours, then neutralized to *p*H 4, and concentrated to dryness *in vacuo*. The viscous product was dissolved in 50 ml. of water and extracted with ether to yield, after distillation, 0.79 g. of substantially pure methylmycarose isovalerate, $n^{25}D$ 1.4510, which was identified through comparison of its infrared absorption spectrum with that of an authentic sample.[5]

The aqueous phase, freed of methylmycarose isovalerate, was concentrated to dryness, dissolved in 20 ml. of 30% hydrochloric acid, and heated under reflux for two hours. The dark solution was filtered from the acid-insoluble tarry residues, washed twice with chloroform, and concentrated to dryness *in vacuo*. The residue was extracted with 10 ml. of hot isopropyl alcohol, adjusted to *p*H 5 with sodium hydroxide, cooled and filtered from the precipitated sodium chloride. The filtrate was concentrated to 4 ml., and 0.1 ml. of water added. On standing overnight, 0.7 g. of colorless crystals separated. Four recrystallizations from hot 96% isopropyl alcohol yielded, after drying at 56°, about 0.2 g. of pure mycaminose hydrochloride monohydrate, m.p. 115–116°, with prior softening when placed in a bath at 90°, temperature rising at 3° per minute; $[\alpha]^{25}D$ +31° (c 1 in water, 24 hours).

Anal. Calcd. for $C_8H_{17}NO_4$·HCl·H_2O: C, 39.10; H, 8.21; N, 5.71; Cl, 14.43; H_2O, 7.34; C-methyl (1), 6.11. Found: C, 38.89; H, 8.01; N, 5.68; Cl, 15.01, H_2O (K.F.) 8.35; C-methyl, 5.42.

Titration shows an equivalent weight of 239 (calcd. 246), pK_b ≈8.5. Mycaminose reduces hot Fehling solution and readily liberates one mole of dimethylamine on hydrolysis in strong alkali.[8] Mycaminose hydrochloride can also be isolated from Magnamycin by this procedure.

Mycaminose Acetates.—The combined mother liquors from the recrystallizations of mycaminose hydrochloride were concentrated to dryness, dissolved in 1 ml. of pyridine and 1 ml. of acetic anhydride, heated on the steam-bath for one hour, then distilled to dryness *in vacuo*. The residue was dissolved in 10 ml. of water, cooled, adjusted to *p*H 10, and extracted with three 10-ml. portions of chloroform.

Distillation of the chloroform extract at 150°, 0.05 mm., yielded 0.7 g. of a viscous oil which slowly crystallized to a solid, m.p. 40–48°. This mixture of triacetates was identical to that obtained on acetylation of pure mycaminose, as judged by melting point, mixed melting point and comparison of infrared spectra.

The mixed triacetates were dissolved in 3 ml. of hot ethanol, and 0.5 g. of picric acid in 2 ml. of hot ethanol added. The yellow crystalline precipitate which formed was filtered from the hot solution, and washed with hot ethanol. The

(7) We are indebted to Dr. Peter P. Regna for this information.

(8) We are indebted to Dr. R. L. Wagner for this information.

crystalline picrate (0.7 g.), m.p. 185–190° dec., was dissolved in 10 ml. of chloroform, and extracted once with 10% sodium hydroxide, then with 1% sodium bicarbonate until free of picric acid.

The chloroform solution was concentrated to dryness to yield colorless crystals which after sublimation weighed 0.2 g., m.p. 93.5–94°, $[\alpha]^{25}D$ +19°.

Anal. Calcd. for $C_{14}H_{25}NO_7$: C, 52.99; H, 7.30; N, 4.42; acetyl (3), 40.68; mol. wt., 317.3. Found: C, 52.94; H, 7.37; N, 4.34; acetyl, 39.15. Titration showed an equivalent weight of 319, $pK_b = 7.4$.

The mother liquors from the triacetylmycaminose picrate, which had precipitated a yellow gum on cooling, were concentrated to dryness, and freed of picric acid as described above. The product, 0.2 g., was sublimed once and recrystallized twice from hexane at −25° to yield 0.05 g. of a pure second isomer of mycaminose triacetate; m.p. 81.2–83°, $[\alpha]^{25}D$ +95°.

Anal. Calcd. for $C_{14}H_{25}NO_7$: C, 52.99; H, 7.30; N, 4.42; acetyl, 40.68. Found: C, 52.75; H, 7.31; N, 4.48; acetyl, 39.49.

The infrared absorption spectra of these isomeric triacetates are distinctive, and indicate that the two preparations are substantially free of one another.

Acknowledgments.—We are indebted to Dr. A. R. English for the antibacterial spectrum data and to Drs. J. F. Gardocki and S. Y. P'an for the toxicity measurements on this compound. We should like to thank Mr. G. B. Hess for spectral measurements, Mr. T. Toolan for the analyses, and Messrs. R. Kersey and F. Leghorn for the microbiological assays. We also thank Drs. R. L. Wagner and P. P. Regna for their interest and suggestions.

BROOKLYN 6, NEW YORK

[CONTRIBUTION FROM THE CENTRAL RESEARCH DEPARTMENT, CROWN ZELLERBACH CORPORATION]

Conidendrin. II.[1] The Stereochemistry and Reactions of the Lactone Ring[2]

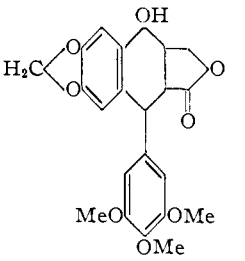
BY MERLE E. CISNEY, WILBUR L. SHILLING, W. M. HEARON AND DAVID W. GOHEEN

RECEIVED APRIL 5, 1954

Dimethyl-α-retrodendrin (III), an isomer of dimethyl-α-conidendrin (II) with the lactone ring disposed as in podophyllotoxin (I), has been synthesized. However, it possessed no tumor-damaging activity. The (−)-α-conidendrin configuration has been demonstrated for III, dimethyl-α-conidendreic acid (VII, "Holmberg's acid") and dimethyl-(+)-isolariciresinol (XII) by the preparation of the C-2 epimer of XII, named dimethyl-β-conidendryl alcohol (XIV).

Podophyllotoxin (I) has been shown to possess tumor-necrotizing action on mouse Sarcoma 37,[3] while dimethyl-α-conidendrin (II) has no such effect. One major difference between these two compounds resides in the position of the lactone ring. In the hope that tumor-necrotizing activity but low toxicity could be obtained, we synthesized the isomer of dimethyl-α-conidendrin in which the lactone ring is reversed as in podophyllotoxin. This compound III which we have designated dimethyl-α-retrodendrin, was devoid of tumor-damaging activity, a result which may possibly be explained on the basis of the steric relationships to be discussed presently.

Omaki,[4] by suitable interconversions, has proved that (−)-α-conidendrin[5] has the same configuration about carbons 2 and 3 as (−)-matairesinol, while Haworth and Kelly[6] have shown that in (−)-matairesinol carbons 2 and 3 have the same absolute configuration. Hence, substituents at carbons 2 and 3 of II must be *trans*. Short heating of II with alcoholic sodium methoxide converts it to the *cis*-lactone, dimethyl-β-conidendrin (IV). This epimerization at C-2 seems to occur through enolization α to the carboxyl group. A completely analogous change of podophyllotoxin produces picropodophyllin, by epimerization about C-3. However, the change in the latter case is effected by much weaker bases, and occurs whenever the lactone ring is opened by alkali. Since the *cis*-lactone, picropodophyllin, does not cause necrosis of mouse sarcoma, it seemed essential that the *trans* relationship of the lactone ring be retained in the preparation of dimethyl-α-retrodendrin. By analogy with the facile epimerization of podophyllotoxin, it appeared likely that dimethyl-α-retrodendrin also would invert readily with base. Later work showed this not to be the case.



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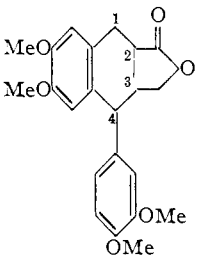
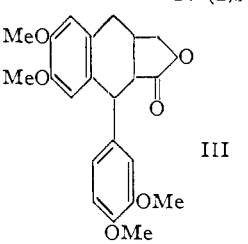


II (2,3-*trans*)
IV (2,3-*cis*)



III

Haworth and Sheldrick[7] reported the synthesis of structure III in several steps, starting with 3,3′,4,4′-

(1) Paper I, W. M. Hearon, H. B. Lackey and W. W. Moyer, THIS JOURNAL, **73**, 4005 (1951).

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

(3) J. Leiter, V. Downing, J. L. Hartwell and H. L. Shear, *J. Natl. Cancer Inst.*, **10**, 1273 (1950).

(4) T. Omaki, *J. Pharm. Soc. Japan*, **57**, 22 (1937).

(5) The designations α and β in the conidendrin series only indicate the *relative* spatial relationship of carbons 2, 3 and 4. However, it is to be understood that all of the derivatives reported in this paper have the same absolute configuration about carbons 3 and 4 as the naturally occurring (−)-α-conidendrin.

(6) R. D. Haworth and W. Kelly, *J. Chem. Soc.*, 384 (1937).

(7) R. D. Haworth and G. Sheldrick, *ibid.*, 636 (1935).