4-Hydroxypretetramids[1,2]

Joseph J. Hlavka, Panayota Bitha, and J. H. Boothe

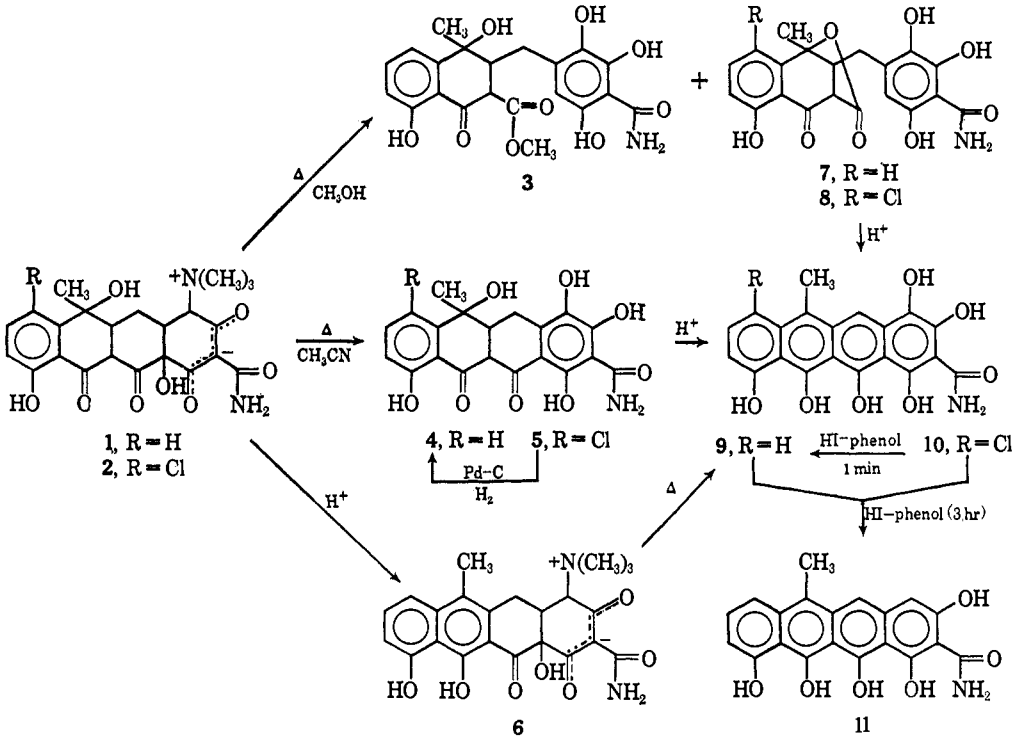
Contribution from the Organic Chemical Research Section, Lederle Laboratories, A Division of American Cyanamid Company, Pearl River, New York 10965.
Received October 3, 1967

Abstract: The conversion of quaternary tetracyclines to the corresponding pretetramids is described. A mechanism for this rearrangement is proposed.

Previous communications from this laboratory describe the synthesis[2] of 4-hydroxy-6-methylpretetramid (**9**) and its important role[3,4] in the biosynthesis of tetracycline. Recently[5] interest has been expressed in related compounds as intermediates in the total synthesis of the tetracyclines.

We now wish to report on an extension of our study which has resulted in the synthesis of the new pretetramids, 4,6-dihydroxypretetramid (**20**) and 7-chloro-4,6-dihydroxypretetramid (**21**), as well as another known biosynthetic intermediate, 4-hydroxypretetramid[6] **17**.

Chart I



Our earlier investigation[2] showed that refluxing tetracycline methyl betaine[7] (**1**) in acetonitrile yielded primarily 4a,12a-anhydro-4-dedimethylamino-4-hydroxytetracycline (**4**). On the other hand, when the betaine **1** was refluxed in methanol we obtained as major products both the methyl ester[8] **3** and the γ-lactone **7**. Reaction of either **4** or **7** with strong acid affords 4-hydroxy-6-methylpretetramid[2-4] (**9**). Alternatively the pretetramid **9** was obtained directly by refluxing anhydrotetracycline methyl betaine (**6**) in acetonitrile.

When 7-chlorotetracycline methyl betaine[7] (**2**) was refluxed in acetonitrile, we isolated the 7-chloro analogs, **5** and **8**. Catalytic hydrogenation of **5** yielded the deschloro isomer **4**. Acid treatment of either derivative **5** or **8** yielded 7-chloro-4-hydroxy-6-methylpretetramid (**10**). Refluxing this material (**10**) for a few minutes in a phenol–hydrogen iodide mixture yielded the deschloro pretetramid **9**; further heating (3 hr) afforded 6-methylpretetramid[6] (**11**) (Chart I).

Using analogous synthetic pathways, we prepared the novel pretetramids, 4,6-dihydroxypretetramid (**20**) and 7-chloro-4,6-dihydroxypretetramid (**21**). Refluxing the methyl betaines **13** or **14** in acetonitrile afforded, respectively, 4a,12a-anhydro-4-dedimethylamino-6-de-

(1) The name "pretetramid" has been suggested for 1,3,10,11,12-pentahydroxynaphthacene-2-carboxamide: J. R. D. McCormick, S. Johnson, and N. O. Sjolander, *J. Amer. Chem. Soc.*, **85**, 1964 (1963).

(2) A preliminary communication on this work was published: J. J. Hlavka, P. Bitha, and J. H. Boothe, *ibid.*, **87**, 1795 (1965).

(3) J. R. D. McCormick, U. H. Joachim, E. R. Jensen, S. Johnson, and N. O. Sjolander, *ibid.*, **87**, 1793 (1965).

(4) J. R. D. McCormick and E. R. Jensen, *ibid.*, **87**, 1794 (1965).

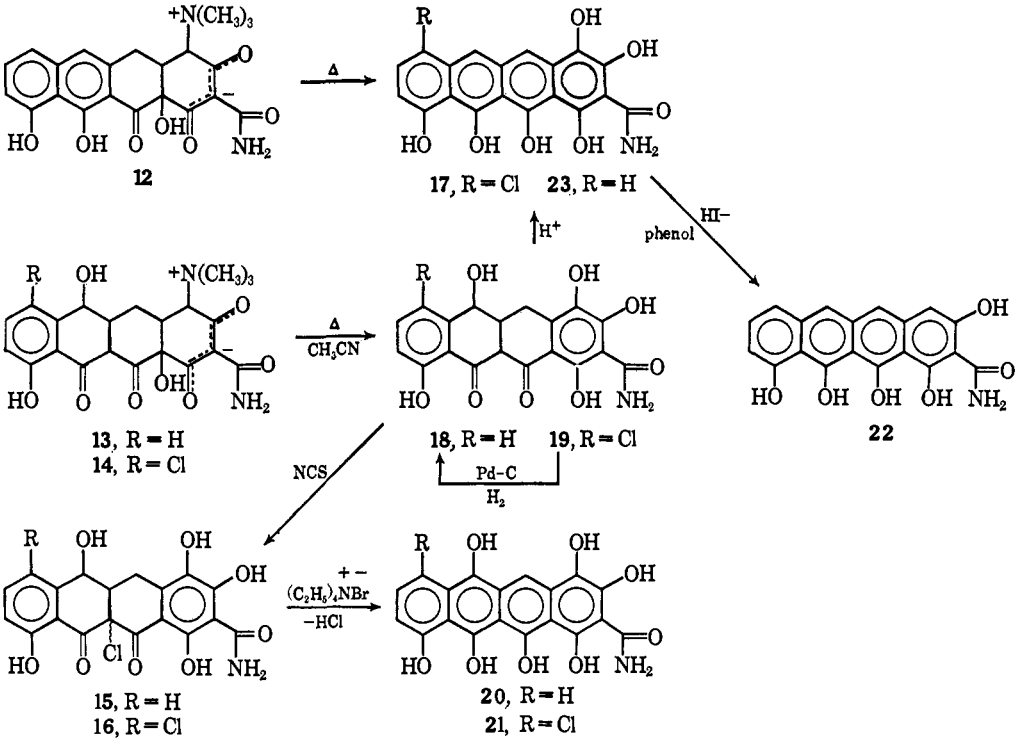
(5) J. E. Baldwin, D. H. R. Barton, L. Bould, and P. D. Magnus, *Chem. Commun.*, 319 (1967).

(6) J. R. D. McCormick in "Biogenesis of Antibiotic Substances," Z. Vanek and Z. Hostalek, Ed., Academic Press Inc., New York, N. Y., 1965, p 73.

(7) J. H. Boothe, G. E. Bonvicino, C. W. Waller, J. P. Petisi, R. W. Wilkinson, and R. B. Broschard, *J. Amer. Chem. Soc.*, **80**, 1964 (1958).

(8) In our previous communication[2] we assigned this material an ε-lactone formulation. Recent mass spectrometric analysis supports the methyl ester structure **3**.

Chart II

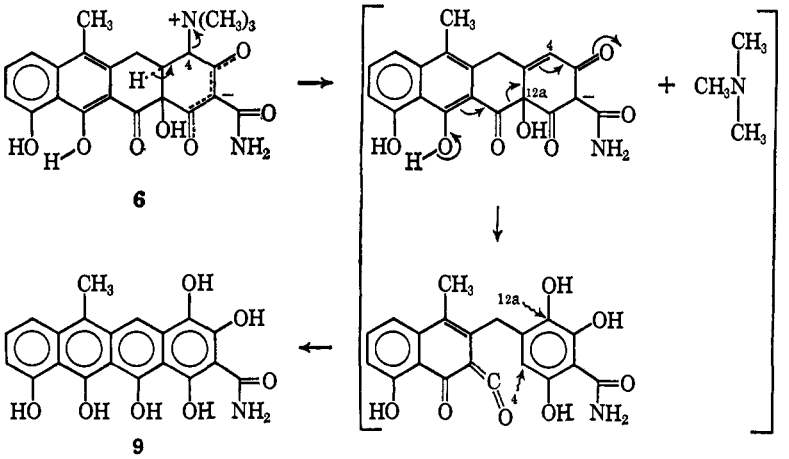


methyl-4-hydroxytetracycline (**18**) or its 7-chloro analog **19**. Molecular weight determinations by mass spectroscopy concur with these formulations. Conversion of **18** and **19** to their 11a-chloro derivatives **15** and **16** followed by dehydrohalogenation[9] yielded the desired pretetramids **20** and **21**. Attempts[10] to biologically convert these pretetramids to new tetracyclines were unsuccessful.

The structure of these pretetramids **20** and **21** was based on composition, their characteristic ultraviolet spectra (as compared to the known 4-hydroxypretetramid),[6] and their conversion to pretetramid (**22**) when reduced with a phenol–hydrogen iodide solution (Chart II).

The 4-hydroxypretetramid **23**, a biosynthetic precursor[6] in the synthesis of 7-chloro-6-demethyltetracycline, was prepared *via* refluxing 6-demethylanhydrotetracycline methyl betaine (**12**) in ethylene glycol monomethyl ether. Another synthetic route to **23** involved conversion of 6-demethyltetracycline methyl betaine (**13**) to **18** with subsequent acid dehydration to yield the 4-hydroxypretetramid **23**. Refluxing either **17**, **20**, **21**, or **23** in a hydrogen iodide–phenol solution afforded the parent pretetramid[6] **22**.

Although a number of possibilities exist for the



mechanism of this rearrangement of the 12a-hydroxyl group to the 4 position (*i.e.*, **1** → **4**, **2** → **5**, **6** → **9**, **13** → **18**, **14** → **19**, and **12** → **23**), we suggest a ring-opening mechanism similar to that formulated by Barton and Scott[11] for the racemization of geodin and by Stork[12] for the racemization of usnic acid.

Experimental Section

All nmr spectra were measured in deuterated dimethyl sulfoxide with tetramethylsilane as the internal standard using a Varian Model A-60 spectrometer. Liquid–liquid partition chromatography was carried out on neutral (acid-washed) diatomaceous earth.

(9) (a) D. N. Kevill, G. A. Coppens, and N. H. Crowell, *J. Org. Chem.*, 28, 567 (1963); (b) J. R. D. McCormick, R. Winterbottom, and P. Bitha, U. S. Patent 3,226,435 (Dec 28, 1965).

(10) J. R. D. McCormick and N. O. Sjolander, Lederle Laboratories Division of American Cyanamid Company, Pearl River, N. Y., private communication.

(11) D. H. R. Barton and A. I. Scott, *J. Chem. Soc.*, 1767 (1958).

(12) G. Stork, *Chem. Ind.* (London), 915 (1955).

4a,12a-Anhydro-4-dedimethylamino-4-hydroxytetracycline (4). A suspension of 2.0 g (4.36 mmol) of tetracycline methyl betaine[7] in 500 ml of acetonitrile was refluxed under a nitrogen atmosphere for 1.5 hr. The heating was stopped, and nitrogen was bubbled through the solution for an additional 15 min until all the trimethylamine vapors had been stripped off. An orange crystalline product separated on cooling and weighed 100 mg; λ_{max} (concentrated H_2SO_4–1% sodium borate) 281, 311, 455, and 520 mμ (log ε 4.22, 4.46, 4.05, and 3.92); mass spectrometric molecular weight 399.

Anal. Calcd for $C_{20}H_{17}NO_8$: C, 60.20; H, 4.26; N, 3.5. Found: C, 59.87; H, 4.45; N, 3.45.

The acetonitrile filtrate was evaporated to a volume of 50 ml and an additional 408 mg of product separated. The second crop was contaminated with a small amount of **7** as determined by infrared spectroscopy.

3-(4-Carbamoyl-2,3,5-trihydroxybenzyl)-1,2,3,4-tetrahydro-4,8-dihydroxy-4-methyl-1-oxo-2-napthoic Acid γ-Lactone (7) and Methyl Ester (3). A suspension of 1.0 g (2.18 mmol) of tetracycline methyl betaine[7] in 160 ml of methanol was refluxed under a nitrogen atmosphere for 1.5 hr. The heat was removed, and nitrogen was bubbled through the solution for an additional 15 min. The solution was evaporated to dryness *in vacuo*. The yellow residue was triturated with heptane leaving 665 mg of crude product. This reaction product was purified by liquid–liquid partition chromatography using the system heptane–ethyl acetate–methanol–water (70:30:17:4). The γ-lactone ($\lambda_{max}^{0.1NHCl}$ 260 and 335 mμ (log ε 4.15 and 3.79); λ_{max}^{KBr} 5.61 μ) was obtained in the 10th hold-back volume (weight 413 mg) and the methyl ester ($\lambda_{max}^{0.1NHCl}$ 261 and 335 mμ (log ε 4.12 and 3.79); λ_{max}^{KBr} 5.75 μ) in the 20th hold-back volume (weight 64 mg). The nmr of the γ-lactone **7** showed an aromatic singlet at τ 3.8 due to the lone proton on the aromatic ring bearing the one carboxamide and three hydroxyl groups.

Anal. Calcd for $C_{20}H_{17}NO_8$ (γ-lactone **7**): C, 60.20; H, 4.26; N, 3.5. Found: C, 60.4; H, 5.0; N, 3.5; mol wt, 388 (mass spectrometric).

Anal. Calcd for $C_{21}H_{21}NO_9$ (methyl ester **3**): C, 58.5; H, 5.02; N, 3.25. Found: C, 58.61; H, 5.36; N, 3.38.

5a,6-Anhydrotetracycline Methyl Betaine (6). Tetracycline methiodide[7] (2.0 g, 3.42 mmol) was dissolved in 25 ml of glacial acetic acid by heating and 5 ml of a 32% solution of hydrobromic acid in acetic acid was added. The resulting solution was heated on a steam bath for 15 min. The reaction mixture was allowed to cool to room temperature for 1 hr and the 5a,6-anhydrotetracycline methiodide crystallized from solution; yield 1.4 g. This material (1.0 g) was suspended in 50 ml of water, and the pH of the mixture was adjusted to 5.1 with 2 *N* sodium hydroxide. The mixture was stirred at room temperature for 1 hr, and the product that separated was filtered and dried; yield 700 mg. This product (100 mg) was recrystallized from 25 ml of ethanol; $\lambda_{max}^{0.1NHCl}$ 272 and 430 mμ (log ε 4.71 and 3.92).

Anal. Calcd for $C_{23}H_{24}N_2O_7 \cdot H_2O$: C, 60.4; H, 5.7; N, 6.13. Found: C, 60.78; H, 5.76; N, 5.85.

4-Hydroxy-6-methylpretetramid (9). Method A. A suspension of 100 mg (0.25 mmol) of **4** in 15 ml of a 32% solution of hydrogen bromide in acetic acid was heated at 50–55° for 3 hr. The mixture was cooled to room temperature and 89 mg of 4-hydroxy-6-methylpretetramid separated; λ_{max} (concentrated H_2SO_4–1% sodium borate) 245, 282, 316, 465, and 525 mμ (log ε 4.09, 4.20, 4.48, 4.11, and 3.97).

Method B. A solution of 100 mg (0.25 mmol) of lactone **7** in 15 ml of a 30% solution of hydrogen bromide in acetic acid was heated for 3 hr at 50–55°. On cooling to room temperature 68 mg of product crystallized.

Method C. A suspension of 200 mg (0.436 mmol) of **6** in 100 ml of acetonitrile is refluxed under a nitrogen atmosphere for 7 hr. The solution was cooled to room temperature and 69 mg of product crystallized.

4a,12a-Anhydro-7-chloro-4-dedimethylamino-4-hydroxytetracycline (5). A suspension of 3.44 g (7.0 mmol) of 7-chlorotetracycline methyl betaine[7] in 630 ml of acetonitrile was refluxed for 1.5 hr under a nitrogen atmosphere. The heating mantle was removed and nitrogen was passed through the solution for an additional 15 min. The solution was evaporated to dryness *in vacuo*, and the residue was triturated with 500 ml of ether. The ether was separated and evaporated to dryness *in vacuo*. The crude residue that remained was purified by partition column chromatography using the system heptane–ethyl acetate–methanol–water (80:20:17:4); yield 59 mg; $\lambda_{max}^{0.1NHCl}$ 250 and 362 mμ (log ε 4.32 and 4.01); mass spectrometric molecular weight, 433.

Anal. Calcd for $C_{20}H_{16}NO_8Cl$: C, 55.30; H, 3.69; N, 3.22. Found: C, 55.55; H, 4.2; N, 3.2.

3-(4-Carbamoyl-2,3,5-trihydroxybenzyl)-1,2,3,4-tetrahydro-5-chloro-4,8-dihydroxy-4-methyl-1-oxo-2-naphthoic acid γ-lactone (8) was obtained from the partition column described in the previous experiment; yield 85 mg; $\lambda_{max}^{0.1NHCl}$ 260 and 352 mμ (log ε 409 and 368); λ_{max}^{KBr} 5.60 μ.

Anal. Calcd for $C_{20}H_{16}NO_8Cl$: C, 55.30; H, 3.69; N, 3.22. Found: C, 55.80; H, 4.40; N, 3.72.

7-Chloro-4-hydroxy-6-methylpretetramid (10). 4a,12a-Anhydro-7-chloro-4-dedimethylamino-4-hydroxytetracycline (**5**) (20 mg, 0.046 mmol) was dissolved in 4 ml of a solution of 32% hydrogen bromide in acetic acid. The solution was evaporated immediately to dryness and the residue was triturated with ether; yield 16.7 mg; λ_{max} (concentrated H_2SO_4–1% sodium borate) 270, 310, and 518 mμ (log ε 4.32, 4.39, and 3.98). For identification this material was converted to both the known 4-hydroxy-6-methylpretetramid (**9**) and the known 6-methylpretetramid (**11**) (see below); mass spectrometric molecular weight of **10**, 415.

Conversion of 7-Chloro-4-hydroxy-6-methylpretetramid (10) to 4-Hydroxy-6-methylpretetramid (11). A solution of 5 mg of the 7-chloro derivative in 2 ml of phenol, 1 ml of 57% hydrogen iodide solution, and one drop of hypophosphorous acid was heated at reflux for 3 hr. On cooling to room temperature overnight 1.6 mg of crystals was obtained identical with 6-methylpretetramid (**11**).

Conversion of 7-Chloro-4-hydroxy-6-methylpretetramid (10) to 4-Hydroxy-6-methylpretetramid (9). A mixture of 10 mg of the 7-chloro derivative in 0.25 ml of phenol and 0.2 ml of 57% hydrogen iodide solution was heated over a flame for 1 min. On cooling a solid crystallized which weighed 7.2 mg and proved to be identical with 4-hydroxy-6-methylpretetramid.

Catalytic Reduction of 4a,12a-Anhydro-7-chloro-4-dedimethylamino-4-hydroxytetracycline (5) to the Deschloro Analog 4. A solution of 10 mg of the 7-chloro derivative **5** in 10 ml of acetonitrile was reduced with hydrogen and 10 mg of a 10% palladium-on-carbon catalyst (at STP) for 16 hr. The catalyst was filtered and the filtrate evaporated to dryness. The residue was identical with **4**.

7-Chloro-6-demethyltetracycline Methyl Betaine. The 7-chloro-6-demethyltetracycline free base (3.0 g) was dissolved in 60 ml of hot tetrahydrofuran. To the clear solution was added 9 ml of methyl iodide, and the mixture was stored at room temperature for 6 days A small amount of solid separated, and this was filtered. To the filtrate was added 200 ml of ether and the methiodide separated; yield 3.18 g. This material was recrystallized from methanol; $\lambda_{max}^{0.1NHCl}$ 228, 270, and 373 mμ (log ε 4.41, 4.14, and 4.03).

The methiodide (200 mg) was dissolved in 5 ml of water and the pH of the solution was adjusted to 6.8 with 20% aqueous sodium acetate. The solid that separated was redissolved by adding 1 ml of methanol. The methanol was then stripped off *in vacuo* and a crystalline product separated; yield 143 mg.

Anal. Calcd for $C_{22}H_{23}N_2O_8Cl \cdot 2H_2O$: C, 51.2; H, 5.23; N, 5.43; Cl, 6.89. Found: C, 51.43; H. 5.02; N, 5.78; Cl, 6.84.

4a,12a-Anhydro-7-chloro-4-hydroxy-6-demethyltetracycline (19). Nitrogen was bubbled through a suspension of 3.0 g of 7-chloro-6-demethyltetracycline methyl betaine (**14**) for 15 min. The suspension was then refluxed under a nitrogen atmosphere for 4 hr. The heat was removed and nitrogen was passed through the solution for an additional 10 min. The small amount of material that did not go into solution was filtered, and the filtrate was evaporated to dryness; yield 2.1 g. This material was recrystallized from 500 ml of methanol; yield 381 mg; $\lambda_{max}^{0.1NHCl}$ 285 and 400 mμ (log ε 3.85 and 4.19); mass spectrometric molecular weight, 419.

Anal. Calcd for $C_{19}H_{14}NO_8Cl$: C, 54.5; H, 3.36; N, 3.34; Cl, 8.46. Found: C, 54.39; H, 3.61; N, 3.43; Cl, 8.72.

7-Chloro-4-hydroxypretetramid (17). A solution of 60 mg of 4a,-12a-anhydro-7-chloro-6-demethyl-4-hydroxytetracycline in 4.0 ml of a solution of 32% hydrogen bromide in acetic acid was stirred at room temperature for 5 min and evaporated to dryness *in vacuo*. The residue was triturated with ether and the solid that remained weighed 58 mg; λ_{max} (concentrated H_2SO_4–1% sodium borate) 285, 318, 481, and 534 mμ (log ε 4.22, 4.61, 4.16, and 3.95); mass spectrometric molecular weight, 401. This material was converted to the known pretetramid (**22**) as follows.

A solution of 10 mg of 7-chloro-4-hydroxypretetramid in 4.0 ml of phenol, 2.0 ml of 57% hydrogen iodide, and two drops of hypophosphorous acid was refluxed for 4 hr. The solution was allowed to cool to room temperature and 2.5 mg of pretetramid[1] (**22**) separated.

6-Demethyltetracycline Methyl Betaine (13). 6-Demethyltetracycline (5 g) was dissolved in 170 ml of hot acetonitrile. To this

solution was added 15 ml of methyl iodide. The resulting solution was stored at room temperature under a nitrogen atmosphere for 7 days and then diluted with 700 ml of ether; yield 5.35 g.

Anal. Calcd for $C_{22}H_{25}N_2O_8I$: I, 22.1. Found: I, 21.6.

The methiodide prepared above (500 mg) was dissolved in 10 ml of methanol, and the pH was adjusted to 6.0 with aqueous sodium hydroxide. The solid (252 mg) that separated was filtered and dried and used directly in the next step below; $\lambda_{max}^{0.1NHCl}$ 269 and 355 mμ (log ε 4.22 and 4.11).

4a,12a-Anhydro-4-dedimethylamino-4-hydroxy-6-demethyltetracycline (18). The 6-demethyltetracycline methyl betaine (prepared above), 2.0 g, was suspended in 500 ml of acetonitrile, and the mixture was refluxed for 2.5 hr under a stream of nitrogen. On cooling, the reaction mixture was filtered, and the filtrate was concentrated to one-half the volume *in vacuo*. The solid that separated weighed 1.2 g. This material was recrystallized from methanol; yield 126 mg; $\lambda_{max}^{0.1NHCl}$ 260 and 480 mμ (log ε 4.13 and 4.27); mass spectrometric molecular weight, 385.

Anal. Calcd for $C_{19}H_{15}NO_8$: C, 59.1; H, 3.92; N, 3.64. Found: C, 59.72; H, 4.71; N, 3.72.

Treatment of this material with 37% hydrogen bromide in acetic acid yielded immediately the known 4-hydroxypretetramid.[6]

4,6-Dihydroxypretetramid (20). A solution of 60 mg of 4a,-12a-anhydro-4-dedimethylamino-4-hydroxy-6-demethyltetracycline and 41.5 mg of N-chlorosuccinimide in 10 ml of ethylene glycol monomethyl ether was stirred at room temperature for 1 hr and evaporated to dryness *in vacuo*. The residue was triturated with ether, and the solid that remained weighed 50 mg. This material (4a,12a-anhydro-11a-chloro-4-dedimethylamino-4-hydroxy-6-demethyltetracycline (**15**)) was dissolved in 6 ml of acetonitrile and to the solution was added 37 mg of tetraethylammonium bromide. The resulting solution was heated at reflux for 30 min. The reaction solution was evaporated to dryness and triturated with ether, and the solid that remained was recrystallized from 2.5 ml of a 57% hydrogen iodide solution to which was added 2.5 ml of melted phenol; yield 26 mg. This material **20** was converted to the known pretetramid **22** by refluxing 4 hr in a mixture of phenol–57% hydrogen iodide; λ_{max} (concentrated H_2SO_4–1% sodium borate) 308, 450, and 500 mμ (log ε 4.31, 3.70, and 3.65).

Anal. Calcd for $C_{19}H_{13}NO_8$: C, 59.7; H, 3.39; N, 3.65. Found: C, 59.53; H, 3.9; N, 3.34.

4a,12a-Anhydro-7,11a-dichloro-4-dedimethylamino-4-hydroxy-6-demethyltetracycline (16). A mixture of 70 mg of N-chlorosuccinimide and 200 mg of 4a,12a-anhydro-7-chloro-4-dedimethylamino-4-hydroxy-6-demethyltetracycline in 50 ml of ethylene glycol monomethyl ether was stirred at room temperature overnight. The solvent was evaporated to dryness *in vacuo* and the residue was triturated with ether; yield 117 mg. The product was crystallized from ether and analyzed as follows.

Anal. Calcd for $C_{19}H_{13}NO_9Cl$: C, 50.2; N, 3.0; Cl, 15.9. Found: C, 50.5; N, 3.6; Cl, 15.9.

7-Chloro-4,6-dihydroxypretetramid (21). A solution of 60 mg of **16** and 54 mg of tetraethylammonium bromide in 15 ml of acetonitrile was refluxed for 15 min, and the solution was allowed to cool to room temperature The solid crystallized; yield 33 mg. This material was recrystallized as follows. A suspension of the material in 0.6 ml of 57% hydrogen iodide and 0.6 ml of melted phenol was heated over a flame for 1 min. On cooling, a solid crystallized which was filtered and washed with ether; yield 27 mg. This material **21** was converted to the known pretetramid **22** by refluxing 4 hr in a mixture of phenol–57% hydrogen iodide, λ_{max} (concentrated H_2SO_4–1% sodium borate) 308, 455 and 525 mμ (log ε 4.66, 4.02 and 3.62).

Anal. Calcd for $C_{19}H_{12}NO_8Cl$: N, 3.35; Cl, 8.49. Found: N, 3.38; Cl, 8.49.

5a,6-Anhydro-6-demethyltetracycline. A solution of 10.0 g of 6-demethyltetracycline in a mixture of 15 ml of 1-butanol and 15 ml of concentrated hydrochloric acid was heated at 75° for 30 min. The resulting solution was stored at room temperature overnight and a solid separated. This was filtered and washed with 50 ml of 2-propanol; yield 10.6 g. A sample (6.5 g) of this material was dissolved in 170 ml of methanol, and the pH of the solution was adjusted to 5.0 with 6 *N* sodium hydroxide. The solid that crystallized weighed 4.8 g.

Anal. Calcd for $C_{21}H_{20}N_2O_7$: N, 6.8. Found: N, 6.63.

5a,6-Anhydro-6-demethyltetracycline Methyl Betaine (12). A solution of 4.7 g of 5a,6-anhydro-6-demethyltetracycline (free base) and 14 ml of methyl iodide was refluxed for 26 hr under a nitrogen atmosphere. The reaction solution was allowed to cool to room temperature and the methiodide crystallized.

Anal. Calcd for $C_{22}H_{23}N_2O_7I$: C, 47.7; H, 4.16; N, 5.05. Found: C, 47.49; H, 4.55; N, 4.79.

A portion of this material (700 mg) was dissolved in methanol and the pH of the resulting solution was adjusted to 5.5 with 6 *N* sodium hydroxide. The solid that crystallized weighed 480 mg; $\lambda_{max}^{0.1NHCl}$ 270 and 425 mμ (log ε 4.77 and 3.98). This material was used without any further purification.

4-Hydroxypretetramid[6] (23). The betaine **12** (3.2 g) prepared above was dissolved in 1 l. of ethylene glycol monomethyl ether; the solution was refluxed under a nitrogen atmosphere for 1 hr. The heat was removed and nitrogen was passed through the solution an additional 15 min. The solution was evaporated to dryness. The residue was crystallized as follows: 280 mg was dissolved in a mixture of 5 ml of 57% hydrogen iodide and 5 ml of melted phenol. The solution was heated over a flame for 1 min and allowed to cool to room temperature. This material proved to be identical with authentic 4-hydroxypretetramid.

Acknowledgments. We wish to thank Dr. A. S. Kende for helpful discussions, Mr. L. Brancone and staff for the analytical data, Mr. G. Morton for the nmr measurements, and Dr. J. Karliner for the mass spectrometric molecular weights.