[CONTRIBUTION FROM THE DEPARTMENT OF CHEMISTRY OF WAYNE UNIVERSITY]

The Partial Structure of Methymycin[1]

BY CARL DJERASSI, A. BOWERS, R. HODGES AND B. RINIKER

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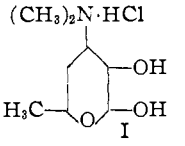
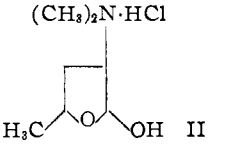
The isolation of the amino sugar desosamine (I) by acid hydrolysis and of 2,4,6-trimethylcyclohex-2-en-1-one (IV) by alkali fusion of methymycin ($C_{28}H_{43}NO_7$) coupled with the location of the lactone termination point accounts for 18 of the 25 carbon atoms of the antibiotic. The nature and location of the hydroxyl groups have been defined precisely and the close relationship to the antibiotics erythromycin and pikromycin is emphasized.

Five of the seven oxygen atoms in the antibiotic methymycin ($C_{28}H_{43}NO_7$)[2] have been shown[3] to be present in the form of an α,β-unsaturated carbonyl function, a lactone (or ester) moiety and two hydroxyl groups. If the presumed[2,3] relationship of methymycin to antibiotics such as pikromycin,[4] erythromycin,[5] magnamycin[6] and narbomycin[7] does indeed obtain, then the remaining two oxygen atoms of methymycin must be present in an acetal linkage involving an amino sugar.

After several unsuccessful attempts, evidence for the presence of an amino sugar was obtained by employing a biphasic, acid hydrolysis system,[8] but insufficient material was isolated for adequate characterization. Finally it was observed that if methymycin was heated for only 10 minutes with 5 *N* hydrochloric acid instead of the more drastic conditions employed earlier,[4-8] over 70% of an amino sugar hydrochloride could be isolated. This was identified as desosamine hydrochloride (I), a cleavage product of a number of antibiotics,[4,5,7] by direct comparison[9] as well as by step-wise oxidation[5] to the pentose II and complete oxidation[5] to crotonaldehyde. The desosamine molecule thus contains one of the two hydroxyl groups of methymycin as well as the dimethylamino function and this raises some interesting questions concerning their behavior on oxidation. It has been reported earlier[3] that chromium trioxide–pyridine oxidation of methymycin apparently converts one of the N-methyl groups to N-formyl, without attacking the two hydroxyl substituents. In fact, the two hydroxyl groups were also resistant to chromium trioxide–sulfuric acid which led to the tentative conclusion that both of them were tertiary. It is clear now (*vide infra*) that only one of them can be tertiary and the unusual behavior of desosamine toward oxidizing agents merits further investigation, which is contemplated when additional supplies become available.



I



II

In the original,[3] small-scale alkali fusion of methymycin, which led to dimethylamine, there was noted the formation of trace amounts of a volatile, pleasant-smelling oil. The isolation and identification of this material has now provided further insight into the structure of methymycin. The fusion with potassium hydroxide was carried out at 360° and the volatile products were distilled directly into a solution of 2,4-dinitrophenylhydrazine. Careful chromatography yielded a 2,4-dinitrophenylhydrazone, $C_{15}H_{18}N_4O_4$, which exhibited an ultraviolet absorption maximum (chloroform solution) at 382 mμ, identical in position and intensity with that observed for the 2,4-dinitrophenylhydrazone of 2-methylcyclohex-2-en-1-one. The unknown degradation product possessed three C-methyl groups (Kuhn–Roth determination) and upon cleavage[10] with hydrochloric acid and stannous chloride afforded the parent ketone $C_9H_{14}O$, whose ultraviolet (λ_{max}^{EtOH} 236 mμ) and infrared spectral properties were only compatible with a cyclohexenone derivative bearing a *single* substituent on the double bond. This was confirmed by hydrogenation to the saturated ketone, whose infrared carbonyl band was typical of a cyclohexanone rather than cyclopentanone derivative. Since the saturated ketone failed to condense with benzaldehyde, this required the presence of methyl groups at both α-positions. These requirements, coupled with the analytical evidence for the presence of three C-methyl groups and the ultraviolet absorption maximum of the ketone and its 2,4-dinitrophenylhydrazone, left for consideration only two structural alternatives, III or IV. The correctness of IV (2,4,6-trimethylcyclohex-2-en-1-one) was established by synthesis involving modified[11] Birch reduction of mesitol methyl ether (V)[12] followed by treatment with acid.

(1) We are greatly indebted to the Squibb Institute for Medical Research for financial support in the form of fellowships and for supplies of methymycin.

(2) M. N. Donin, J. Pagano, J. D. Dutcher and C. M. McKee, "Antibiotics Annual 1953–1954," Medical Encyclopedia, Inc., New York, N. Y., p. 179.

(3) C. Djerassi, A. Bowers and H. N. Khastgir, THIS JOURNAL, **78**, 1729 (1956).

(4) H. Brockmann and W. Henkel, *Ber.*, **84**, 284 (1951); H. Brockmann and R. Oster, *Naturwiss.*, **42**, 155 (1955), and references cited therein.

(5) E. H. Flynn, M. V. Sigal, P. F. Wiley and K. Gerzon, THIS JOURNAL, **76**, 3121 (1954); R. K. Clark, Jr., *Antibiotics & Chemotherapy*, **3**, 663 (1953).

(6) R. L. Wagner, F. A. Hochstein, K. Murai, N. Messina and P. P. Regna, THIS JOURNAL, **75**, 4684 (1953); F. A. Hochstein and P. P. Regna, *ibid.*, **77**, 3353 (1955).

(7) R. Corbaz, L. Ettlinger, E. Gaumann, W. Keller, P. Kradolfer, E. Kyburz, L. Neipp, V. Prelog, R. Reusser and H. Zahner, *Helv. Chim. Acta*, **38**, 935 (1955).

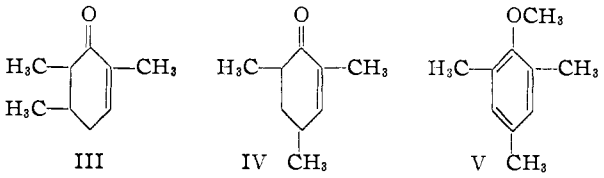
(8) We are indebted to Prof. R. B. Woodward (Harvard University) for the experimental details; *cf.* P. F. Wiley, K. Gerzon, E. H. Flynn, M. V. Sigal and U. C. Quarck, THIS JOURNAL, **77**, 3676 (1955).

(9) We are grateful to Drs. R. G. Jones and T. P. Carney (Eli Lilly and Co.) for a specimen of desosamine hydrochloride obtained from erythromycin (ref. 5).

(10) The conditions of J. Demaecker and R. H. Martin (*Nature*, **173**, 266 (1954)) had to be modified somewhat, since they are not applicable directly to simple alicyclic ketones.

(11) A. L. Wilds and N. Nelson, THIS JOURNAL, **75**, 5360 (1953).

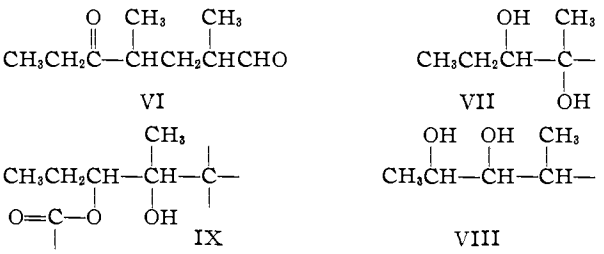
(12) K. v. Auwers, *Ann.*, **415**, 156 (1918).



III IV V

While the isolation of 2,4,6-trimethylcyclohex-2-en-1-one (IV) accounts for nine carbon atoms of the methymycin molecule, it is clear (on spectroscopic and other grounds[3]) that this fragment *per se* is not present in the antibiotic but rather is formed by a cyclization and cleavage process. The simplest, though not necessarily the only formulation would involve base-catalyzed cyclization of a keto-aldehyde VI, but since neither a saturated keto nor aldehyde group is present in methymycin,[2,3] it would be necessary to rationalize the formation of the hypothetical precursor VI in the alkali fusion. The ketonic function of VI could have been produced in the high temperature fusion either by oxidation of a secondary alcohol or by thermal dehydration of a glycol. The first alternative is unlikely, since oxidation experiments[3] with methymycin preclude the presence of an easily oxidizable, secondary alcohol and it will be necessary, therefore, to consider the second possibility.

Theoretically, two glycol fragments, represented by VII and VIII, could give rise to the keto group of VI, but since methymycin is unattacked by periodic acid,[3] one of the two hydroxyl groups must be blocked as a lactone (or ester) which is known to be present in the antibiotic. In view of the fact that irrespective of which hydroxyl group of VIII is assumed to be involved in lactone formation, the remaining one would have to be secondary (and hence readily oxidized by chromium trioxide), only the partial structure IX is in accordance with the experimental facts. This could be verified readily, since lithium aluminum hydride reduction of IX should give rise to the free glycol grouping (VII) which should now be attacked by periodic acid. Indeed, when the crude lithium aluminum hydride reduction product of methymycin was treated with periodic acid, one equivalent of the reagent was consumed rapidly and propionaldehyde could be isolated in good yield.



VI VII

IX VIII

The above reaction sequence also opened an experimental approach to determine which oxygen function was lost in tetrahydrodesoxymethymycin,[3] the hydrogenolysis product of methymycin. It had been pointed out earlier[3] that the oxygen atom removed under those conditions must have been one of the two hydroxyl groups originally present in methymycin rather than the carbonyl group. Since the position of the two hydroxyl groups has now been defined, one of them forming part of the desosamine fragment (I) and the other being located next to the lactone (IX), it remained only to determine which fragment (I or IX) had been altered in tetrahydrodesoxymethymycin. Acid hydrolysis of tetrahydrodesoxymethymycin again led to desosamine hydrochloride (I), but lithium aluminum hydride reduction now afforded a product which was not affected by periodic acid. It follows that the tertiary hydroxyl group in IX had been removed by hydrogenolysis.[13]

The isolation of desosamine hydrochloride (I) and the demonstration of the presence of the fragment IX in methymycin establish the nature of 14 out of the 25 carbon atoms of methymycin. The carbon fragment IX is also present in erythromycin[14] and in pikromycin[4] and there is no question now of the close structural relation between methymycin and these antibiotics. The above-mentioned experiments and the resemblance to pikromycin and erythromycin, in which the presence of a large lactone ring has been demonstrated, require that a similar lactone ring[15] be present in methymycin.

Whether trimethylcyclohexenone (IV) arose from a hypothetical intermediate such as VI or whether it comes from the remaining and as yet unidentified portion of the methymycin molecule will become apparent from experiments on a desosamine-free cleavage product to be described in a future publication. It should be noted that the remaining 11 carbon atoms must contain the α,β-unsaturated carbonyl system,[2,3] the termination point of the lactone ring (IX) and the point of attachment of the desosamine moiety (I).

Experimental[16]

Isolation of Desosamine Hydrochloride (I).—A solution of 2.0 g. of methymycin in 50 cc. of 5 *N* hydrochloric acid was heated on the steam-bath for 10 minutes and then extracted with chloroform. The aqueous layer was evaporated to dryness *in vacuo* and the residue was crystallized from methanol–acetone to yield 0.663 g. of crude desosamine hydrochloride, m.p. 175–185°. Two recrystallizations from the same solvent pair led to the analytical sample, m.p. 187–189° dec., undepressed upon admixture with an authentic specimen,[9] $[\alpha]_D$ +50.5° (water), positive iodoform and Fehling tests.

Similar hydrolysis of tetrahydrodesoxymethymycin[3] yielded desosamine hydrochloride, m.p. 187–189° dec., $[\alpha]_D$ +51° (water), the infrared spectrum (potassium bromide pellet) of which was superimposable with that of the authentic material.[9]

Anal. Calcd. for $C_8H_{17}NO_3 \cdot HCl$: C, 45.40; H, 8.59; N, 6.63; Cl, 16.78; 2N-CH_3, 14.2; neut. equiv., 211.7. Found: C, 45.14; H, 8.53; N, 6.82; Cl, 17.38; N-CH_3, 13.03; neut. equiv. ($HClO_4$ titration), 205.

For further characterization, desosamine hydrochloride was oxidized with one equivalent of sodium metaperiodate to the pentose hydrochloride (II),[5] m.p. 167–169° (positive Fehling reaction) and with an excess of this reagent to

(13) The reduction experiments reported earlier (ref. 3) suggest that the presence of the carbonyl group is essential for hydrogenolysis. Since the carbonyl group cannot be located α or β to the hydroxyl function in IX, it is possible that some type of *trans*-annular activation is responsible.

(14) P. F. Wiley, K. Gerzon, E. H. Flynn, M. V. Sigal and U. C. Quarck, THIS JOURNAL, **77**, 3677 (1955).

(15) Additional circumstantial evidence is given in ref. 3.

(16) All melting points were determined on the Kofler block. We are indebted to Mrs. Dolores Phillips for the infrared and ultraviolet spectral determinations and to Mr. Joseph F. Alicino (Squibb Institute for Medical Research) for the microanalyses.

crotonaldehyde,[5] which was identified as the 2,4-dinitrophenylhydrazone.

Alkali Fusion of Methymycin.—A mixture of 4.0 g. of methymycin and 8.0 g. of powdered potassium hydroxide was placed in a Wood metal-bath at 200° and the temperature raised over a period of 5 minutes to 360°. Heating was continued at 360° for an additional 15 minutes and the volatile material was distilled directly into a solution of 700 mg. of 2,4-dinitrophenylhydrazine in 40 cc. of methanol containing a few drops of concd. hydrochloric acid. The fusion mixture was cooled, dissolved in water, extracted with ether and the organic phase was washed with dilute hydrochloric acid, water, dried and evaporated. The residue, dissolved in methanol, was added to the above dinitrophenylhydrazine solution which was then refluxed for 15 minutes. After cooling and dilution with water, isolation with benzene afforded a product which was dissolved in benzene and filtered through 15 g. of Merck acid-washed alumina in order to separate unreacted reagent which remained on the column. The resulting hydrazone mixture (400 mg.) was then chromatographed in hexane–benzene (1:1) solution on 25 g. of alumina. Elution with the same solvent mixture afforded a fraction (160 mg.), which crystallized from hexane as needles, m.p. 145–163°. Further chromatography and several recrystallizations from hexane afforded the analytical sample of 2,4,6-trimethylcyclohex-2-en-1-one (IV) 2,4-dinitrophenylhydrazone, m.p. 168–171°, $\lambda_{max}^{CHCl_3}$ 382 mμ, log ε 4.38.[17]

Anal. Calcd. for $C_{15}H_{18}N_4O_4$: C, 56.59; H, 5.70; N, 17.60; 3C-CH_3, 14.15. Found: C, 56.75; H, 5.98; N, 17.68; C-CH_3,[18] 15.51, 16.71.

Further elution with benzene–hexane afforded 145 mg. of a fraction, which crystallized from methanol as needles, m.p. 134–142°. Repeated chromatography and recrystallization raised the m.p. to 146–153°; judging from the analysis, ultraviolet ($\lambda_{max}^{CHCl_3}$ 358 mμ) and infrared spectra, the substance appears to be slightly contaminated propionaldehyde 2,4-dinitrophenylhydrazone.[19] No further work was done with this material.

Anal. Calcd. for $C_9H_{10}N_4O_4$: C, 45.38; H, 4.23; N, 23.52. Found: C, 46.05; H, 4.05; N, 22.70.

In view of the very limited amounts of 2,4,6-trimethylcyclohex-2-en-1-one 2,4-dinitrophenylhydrazone obtained from the alkali fusion, most of the succeeding experiments were carried out on a very small scale, using spectral measurements for characterization purposes. As pointed out in the Discussion, this information was sufficient to reduce the structural possibilities to only two (III and IV) which could then be settled by synthesis.

Model experiments with 2-methylcyclohex-2-en-1-one dinitrophenylhydrazone[17] showed that Demaecker and Martin's[10] procedure for the cleavage of dinitrophenylhydrazones would have to be modified (shorter reaction time, reduced amount of acid, higher dilution) in order to avoid the formation of condensation products, and the following method was employed for the methymycin degradation product.

A 25-mg. sample of the 2,4-dinitrophenylhydrazone in 25 cc. of acetone containing 0.625 cc. of concd. hydrochloric acid was refluxed for 45 minutes, 750 mg. of stannous chloride in 0.75 cc. of concd. hydrochloric acid and 1.5 cc. of water was added and heating was continued for 35 minutes under nitrogen. After dilution with water, the product was isolated with ether and represented a mobile liquid with an odor very similar to that of 2-methylcyclohex-2-en-1-one; λ_{max}^{EtOH} 236 mμ (log ε 4.20 assuming a 100% yield in the cleavage), $\lambda_{max}^{CCl_4}$ 5.94 μ. The unsaturated ketone IV was hydrogenated for 2.5 hours in 20 cc. of ethanol with 13 mg. of 5% palladized charcoal catalyst and the crude product (containing some derived alcohol by infrared determination) was oxidized with 8 *N* chromium trioxide reagent. The product was isolated with ether and distilled at 100° and 25 mm. A carbon tetrachloride solution of the ketone exhibited a sharp band at 5.82 μ; under the same conditions, 4-methylcyclohexanone showed a band at 5.82 μ, while cyclopentanone had one at 5.68 μ.

The above saturated ketone was dissolved in 0.1 cc. of ethanol and mixed with 0.05 cc. of 10% sodium hydroxide solution and 0.01 cc. of benzaldehyde. The homogeneous solution was left at room temperature overnight, ether was added and the organic layer was washed thoroughly with sodium bisulfite and with water. The residue, still possessing an odor of benzaldehyde, was passed in hexane solution through 1 g. of alumina; the eluted product did not exhibit any high selective absorption in the ultraviolet indicating that no condensation had occurred with benzaldehyde.

Synthesis of 2,4,6-Trimethylcyclohex-2-en-1-one (IV) from Mesitol Methyl Ether (V).—A solution of 3.3 g. of mesitol methyl ether[12] in 200 cc. of propylene glycol monomethyl ether was added with stirring to 300 cc. of liquid ammonia cooled to −50°. Lithium ribbon (15 g.) was slowly added and after 2 hours the blue color had disappeared. Ammonium chloride (150 g.) was added to the solution and the ammonia was allowed to evaporate. After addition of water, the crude product was isolated with ether and directly cleaved with acid by refluxing for 40 minutes with 25 cc. of 10% hydrochloric acid and 50 cc. of methanol. The resulting product, separated by ether extraction, was mixed with a solution of *ca.* 3 g. of 2,4-dinitrophenylhydrazine in 25 cc. of methanol and 4 cc. of concd. hydrochloric acid. After 10–15 minutes, the product was extracted with benzene and chromatographed as indicated above for the methymycin degradation product; yield 1.15 g., m.p. 155–160°. The pure sample, after recrystallization from methanol and from hexane exhibited m.p. 167.5–170.5°, undepressed upon admixture with the methymycin degradation product, $\lambda_{max}^{CHCl_3}$ 382.5 mμ, log ε 4.43; the infrared spectra of the two specimens were identical.

Isolation of Propionaldehyde from Methymycin and Tetrahydromethymycin.—A solution of methymycin (1.75 g.) in 200 cc. of ether was added to 3.0 g. of lithium aluminum hydride in 100 cc. of ether and the resulting mixture was refluxed for 10 hours. The excess reagent was decomposed with ethyl acetate, aqueous saturated sodium sulfate was added to precipitate inorganic salts and this was followed by the addition of anhydrous sodium sulfate. The mixture was filtered, the residue was extracted four times with chloroform, the combined solutions were evaporated to dryness and codistilled once with toluene to remove all volatile material; yield 1.80 g. of a gum, which did not exhibit any carbonyl absorption in the infrared.

The above gum (1.5 g.) was treated with 2.47 g. of periodic acid in 100 cc. of water, one equivalent of periodic acid having been consumed within 45 minutes after which time no more reagent was taken up.[20] The resulting solution was steam distilled into a solution of 2,4-dinitrophenylhydrazine hydrochloride in methanol. After processing in the usual manner, including chromatography on alumina, there was obtained 0.57 g. of propionaldehyde 2,4-dinitrophenylhydrazone; after recrystallization from methanol m.p. and mixture m.p. 145–150°, the wide range presumably being due to a mixture of forms.[19] Identity was further established by infrared comparison.

Anal. Calcd. for $C_9H_{10}N_4O_4$: C, 45.31; H, 4.23; N, 23.52. Found: C, 45.38; H, 4.23; N, 23.52.

In a second experiment, 0.31 g. of the lithium aluminum hydride reduction product was again oxidized with periodic acid and the reaction mixture was distilled into a solution of 300 mg. of dimedone in 10 cc. of ethanol. After standing overnight at 0°, 95 mg. of the derivative (m.p. 148–152°) separated, raised by recrystallization from ethanol to 153–154.5°. Identity with the corresponding derivative of propionaldehyde was established by mixture melting point determination and infrared comparison.

Tetrahydromethymycin[3] and tetrahydrodesoxymethymycin[3] were treated under identical conditions with lithium aluminum hydride and then periodic acid. The former yielded propionaldehyde while the reduction product of tetrahydrodesoxymethymycin was not affected by periodic acid.

Acknowledgment.—We should like to acknowl-

(17) The 2,4-dinitrophenylhydrazone of 2-methylcyclohex-2-en-1-one, prepared according to W. W. Rinne, H. R. Deutsch, M. I. Bowman and I. B. Joffe (THIS JOURNAL, **72**, 5759 (1950)) exhibited λ_{max}^{CHCl} 382 mμ, log ε 4.40.

(18) The Kuhn–Roth determination in such cases apparently gives somewhat high values as exemplified with the known (ref. 17) dinitrophenylhydrazone of 2-methylcyclohex-2-en-1-one which was run at the same time (*Anal.* Calcd. for $C_{15}H_{14}N_4O_4$: C-CH_3, 5.17. Found: C-CH_3, 6.67).

(19) *Cf.* G. L. Clark, W. I. Kaye and T. D. Parke, *Ind. Eng. Chem., Anal. Ed.*, **18**, 310 (1946).

(20) Methymycin was treated under identical conditions at the same time; no reagent uptake was observed after two hours (*cf.* ref. 3).

edge the benefit of a stimulating discussion with Professor R. B. Woodward (Harvard University) and with Dr. K. Gerzon (Eli Lilly and Co.).

DETROIT, MICHIGAN

[CONTRIBUTION FROM THE CHEMICAL RESEARCH AND DEVELOPMENT DIVISION OF THE SCHERING CORPORATION]

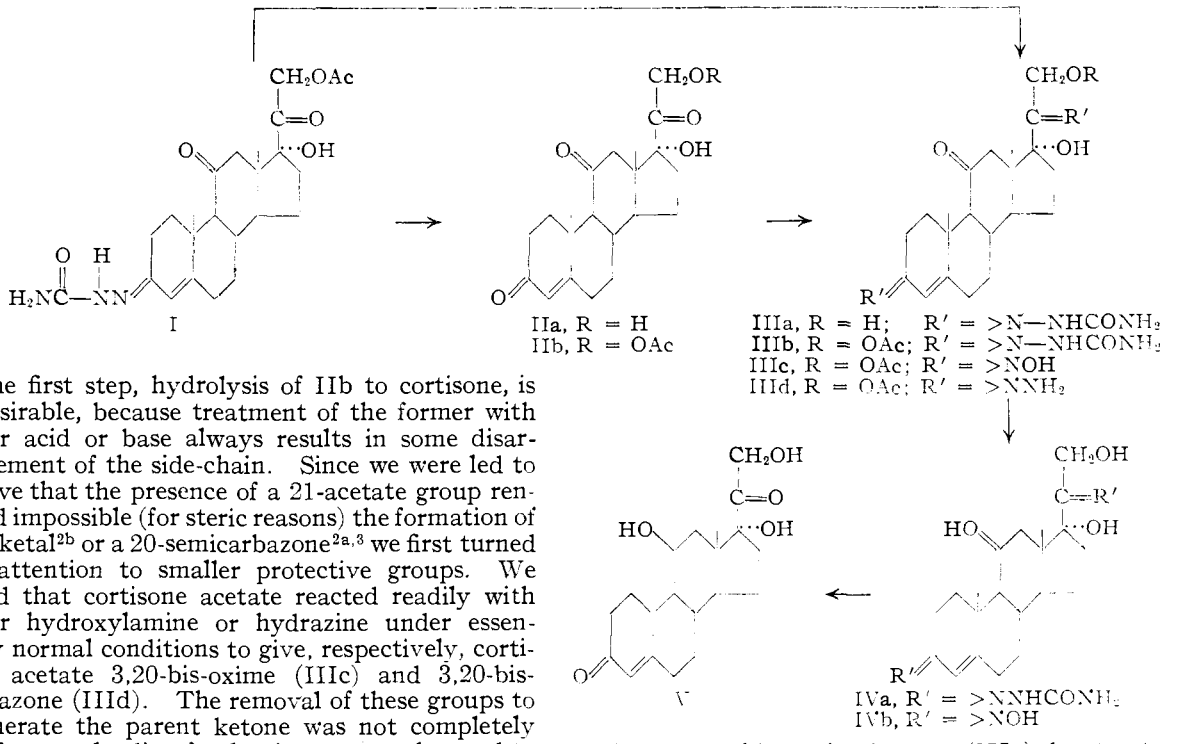
11-Oxygenated Steroids. XVI. The Preparation of Hydrocortisone from Cortisone Acetate[1]

BY EUGENE P. OLIVETO, RICHARD RAUSSER, LOIS WEBER, ELLIOT SHAPIRO, DAVID GOULD AND E. B. HERSHBERG

RECEIVED NOVEMBER 7, 1955

Cortisone acetate (IIb) has been converted into its 3,20-bis-oxime, bis-hydrazone and bis-semicarbazone. The latter, upon reduction with potassium borohydride followed by cleavage of the semicarbazone groups with nitrous acid, gave hydrocortisone (V) in 65–70% over-all yield from IIb.

The published[2] syntheses of hydrocortisone (V) from cortisone acetate (IIb) involve hydrolysis of the latter to cortisone, selective protection of the C-3 and C-20 carbonyls by semicarbazone[2a] or ketal[2b] formation, reduction of the 11-ketone by a suitable metal hydride, and removal of the protective groups. Although these processes give low yields, the ready availability of IIb made attractive a re-investigation of its conversion to V.



The first step, hydrolysis of IIb to cortisone, is undesirable, because treatment of the former with either acid or base always results in some disarrangement of the side-chain. Since we were led to believe that the presence of a 21-acetate group rendered impossible (for steric reasons) the formation of a 20-ketal[2b] or a 20-semicarbazone[2a,3] we first turned our attention to smaller protective groups. We found that cortisone acetate reacted readily with either hydroxylamine or hydrazine under essentially normal conditions to give, respectively, cortisone acetate 3,20-bis-oxime (IIIc) and 3,20-bis-hydrazone (IIId). The removal of these groups to regenerate the parent ketone was not completely satisfactory, lending further impetus to the need to form the 3,20-bis-semicarbazone of IIb.

When IIb was refluxed overnight in aqueous methanol with *unbuffered* semicarbazide hydrochloride a product was obtained, in modest yield, which was not identical with cortisone acetate 3-monosemicarbazone or cortisone 3,20-bis-semicarbazone. Its nitrogen content indicated the presence of two semicarbazone groups, and its infrared spectrum disclosed the retention of the 21-acetate group but the loss of the 20-carbonyl group. This new compound was therefore formulated as cortisone acetate 3,20-bis-semicarbazone (IIIb). This same material also could be obtained from cortisone 3,20-bis-semicarbazone (IIIa) by treatment with acetic anhydride and pyridine at room temperature. The use of pyridine as solvent also gave IIIb, but the best yields (99–100%) were obtained in aqueous methanol upon the addition of a small amount of pyridine.[4] Semicarbazide base

(1) Paper XV, H. L. Herzog, C. C. Payne, M. E. Tully, M. A. Jevnik, E. B. Hershberg, A. Nobile, W. Charney, C. Federbush, D. Sutter and P. L. Perlman.

(2) (a) N. L. Wendler, Huang-Minlon and M. Tishler, *ibid.*, **73**, 3818 (1951); (b) R. Antonucci, S. Bernstein, M. Heller, R. Lenhard, R. Littell and J. H. Williams, *J. Org. Chem.*, **18**, 70 (1953).

(3) O. Mancera, THIS JOURNAL, **72**, 5752 (1950); G. Fleisher and E. C. Kendall, *J. Org. Chem.*, **16**, 556 (1951).

(4) The use of pyridine and semicarbazide hydrochloride in the formation of semicarbazones has been noted before [I. Hopper, *J. Roy. Tech. Coll. Glasgow*, **2**, 52 (1929); *C. A.*, **23**, 3903 (1929)]. Using this technique H. Reich and B. Samuels [*J. Org. Chem.*, **19**, 1041 (1954)] prepared 21-acetoxypregnenolone semicarbazone, but they did not attempt to prepare the semicarbazone of a 20-keto-17α-hydroxy-21-acetoxysteroid.