on the quinone ring (no adjacent proton) in both I-A and II-A.

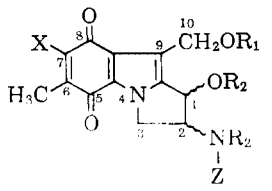
II 

TABLE B

	X	Z	R_1	R_2
II-A	H_3CO	H	$CONH_2$	H
II-B	H_3CO	CH_3	$CONH_2$	H
II-F	HO	H	$CONH_2$	H
II-G	H_3CO	H	$CONH_2$	$COCH_3$
II-H	H_3CCO_2	H	$CONH_2$	$COCH_3$
II-J	HO	CH_3	$CONH_2$	H
II-K	H_3CCO_2	CH_3	$CONH_2$	$COCH_3$
II-L	HO	H	H	H
II-M	H_3CCO_2	H	$COCH_3$	$COCH_3$

The foregoing data suffice to establish the interrelations and chromophores of the antibiotics in this series; the following communication[17] presents additional data sufficient to prove the total structures.

(17) J. S. Webb, D. B. Cosulich, J. H. Mowat, J. B. Patrick, R. W. Broschard, W. E. Meyer, R. P. Williams, C. F. Wolf, W. Fulmor, C. Pidacks and J. E. Lancaster, *J. Am. Chem. Soc.*, **84**, 3187 (1962).

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RECEIVED JUNE 27, 1962

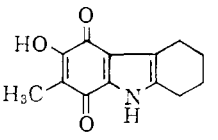
THE STRUCTURES OF MITOMYCINS A, B, AND C AND PORFIROMYCIN—PART II

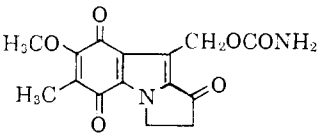
Sir:

In the preceding communication[1] we report the interrelations and chromophores of the mitomycins and porfiromycin. In the determination of the rest of the skeletal structure and the arrangement of functional groups, the reactions described below were of major significance.

apo-Mitomycin A (II-A)[2] formed a diacetyl derivative (II-G) m.p. 244–246.5° dec., with carbonyl bands at 6.44 μ (amide II) and 5.75 μ (alkyl acetate) suggesting the presence in II-A of alkyl NH_2 and OH, neither of which was present in I-A. One of the two remaining nitrogen atoms and both unassigned oxygen atoms in II-A and II-F were part of a $—OCONH_2$ group: hydrolysis of either II-A or II-F (and even I-A) in 6 *N* HCl at 25° produced one mole each of CO_2 and NH_4^+ and a new compound (II-L) [$C_{13}H_{14}N_2O_5$; $[\alpha]^{25}{}_{6907}$ + 26 ± 5° (1% 0.1 *N* HCl); tetraacetyl derivative (II-M) m.p. 225–230°], but in strong base neither NH_3 nor $CO_3^=$ were formed from II-A and II-F until after brief acidification.[3] The band at 5.8 μ, absent in II-L but present in I-A, -B, -C and -D; II-A, -B, -F and -J, was attributed to a carbamate carbonyl.[4] By exclusion the remaining nitrogen atom in II-A must have been the original nitrogen of the aminobenzoquinone in I-A; in II-A it was neither hydrolyzable nor basic; there remained the possibility that it was heterocyclic. In fact the ultraviolet spectrum of 5,6,7,8-tetrahydro-3-hydroxy-2-methyl-1,4-carbazoledione[5] (III) [$\lambda_{max}^{0.1\ N\ HCl}$ 237 mμ (ϵ 20,200), 293 mμ (ϵ 19,000), 370 mμ (ϵ 4,330), 510 mμ (ϵ 1,390), $\lambda_{max}^{0.1\ N\ NaOH}$ 246 mμ (ϵ 24,500), 306 mμ (ϵ 12,500), 365 mμ (ϵ 4,620), 595 mμ (ϵ 1,610)], was nearly identical with that of II-F.

The arrangement of the groups (—OH, $—NH_2$, $—CH_2OCONH_2$) on the skeleton of II-A was established by treatment of it with HNO_2 to produce a compound (IV) $C_{15}H_{14}N_2O_6$, which contained a new carbonyl group, λ_{max}^{KBr} 5.77 μ, was optically inactive, and lacked the NH_2 and OH groups of IIA. The ultraviolet spectrum of IV [$\lambda_{max}^{CH_3OH}$ 280 mμ (ϵ 41,400)] indicated probable conjugation of the new carbonyl with the indoloquinone chromophore, but was *unlike* that of ethyl 5-hydroxy-2,6-dimethyl-4,7-dioxo-3-indolecarboxylate[6]; therefore, the new carbonyl probably was not at the 3-indole position and hence must be at 2-. The n.m.r. spectrum of IV (D_6-dimethyl sulfoxide) displayed two widely separated triplets characteristic of an A_2X_2 pattern not shown by II-A. This suggested the presence of the moiety $Y—CH_2—CH_2—CO—$ at the 2-indole position. Y had to be the indole nitrogen: the $—CH_2OCONH_2$ group in IV must be at the 3- rather than 1-indole position since strong acid hydrolysis of IV produced no formaldehyde, and acid permanganate oxidation of IV yielded β-alanine, identified by paper chromatography[7]


III


IV

(1) J. S. Webb, D. B. Cosulich, J. H. Mowat, J. B. Patrick, R. W. Broschard, W. E. Meyer, R. P. Williams, C. F. Wolf, W. Fulmor, C. Pidacks and J. E. Lancaster, *J. Am. Chem. Soc.*, **84**, 3185 (1962).

(2) All formula rubrics refer to Tables A and B in the preceding Communication.

(3) T. W. J. Taylor and W. Baker, "Sidgwick's Organic Chemistry of Nitrogen," Oxford University Press, London, England, 1942, p. 272.

(4) S. Pinchas and D. Ben-Ishai, *J. Am. Chem. Soc.*, **79**, 4099 (1957).

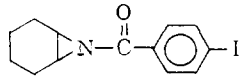
(5) Prepared by Dr. W. Remers of these Laboratories, unpublished data.

(6) H. Teuber and G. Thaler, *Ber.*, **91**, 2253 (1958).

(7) E. D. Moffat and R. I. Lytle, *Anal. Chem.*, **31**, 926 (1959).

and by microbiological assay.[8] It was now clear that IV must have been formed by a pinacolic deamination of a 1,2-aminoalcohol function in II-A; indeed reaction of II-A with phosgene[9] yielded a new compound with a carbonyl, λ_{max}^{KBr} 5.68 μ, characteristic of an oxazolidinone.[4]

The formation by acid hydrolysis of a 1,2-amino alcohol in II-A from a group in the precursor I-A containing only $\rangle$NH suggested that I-A must contain a fused ring aziridine structure. The presence of this unusual group was supported by the following spectral properties: (1) in antibiotics I-A, -B, -C and -D: weak bands in the 3.3 μ region assignable to aziridine C-H stretching,[10] (2) in N-acetyl I-A (prepared *via* 1-acetylimidazole),[11] a carbonyl band at 5.84 μ,[12] (3) in N-(4-iodobenzoyl) 1-A (also prepared *via* 1-acylimidazole)[11] a new λ_{max} at 262 mμ (in addition to the λ_{max} found in I-A) not in the usual location for *p*-iodobenzamide (approx. 252 mμ) but in good agreement with the λ_{max} of 1-(4′-iodobenzoyl)-1-azabicyclo[4.1.0]heptane (V), m.p. 124–5°, prepared similarly.[11]



V

Finally, the acid-labile methoxyl group (6.8 τ, $CDCl_3$) could be assigned to either position C-9 or C-9a in structure I-A. A detailed analysis of the n.m.r. spectrum[13] of I-A clearly indicated 9a; a typical 12 peak ABX pattern was present as expected for the coupling of a single proton on the asymmetric C-9 with the two dissimilar protons at C-10. The spectrum of I-A did not have the doublet and non-equivalence quartet that would be expected if there were a methoxyl at 9 and a proton at 9a. In fact all details[14] of the n.m.r. spectra of I-A, -B, -C and -D are in complete accord with the proposed structures.

After the completion of this work a complete 3-dimensional X-ray structure analysis, carried out by Dr. A. Tulinsky on the N-4′-bromobenzenesulfonyl derivative, m.p. 130–145°, of I-A (prepared[15] by us specifically for this purpose) confirmed our proposed structure in every respect and in addition revealed the relative stereochemistry at the four asymmetric centers, 1, 2, 9, 9a. His work is reported in an accompanying Communication.[16]

(8) H. P. Sarett and V. H. Cheldelin, *J. Bacteriology*, **49**, 31 (1945).

(9) G. M. Tener and H. G. Khorana, *J. Am. Chem. Soc.*, **79**, 437 (1957). The yield of this cyclic derivative was nearly 50% indicating at least this proportion of *cis*-isomer in the mixture; see (13) in previous communication.

(10) H. T. Hoffman, Jr., *et al.*, *ibid.*, **73**, 3028 (1951); there were no aromatic or ethylenic protons in the n.m.r. spectra of the antibiotics.

(11) G. W. Anderson and R. Paul, *ibid.*, **80**, 4423 (1958).

(12) H. C. Brown and A. Tsukamoto, *ibid.*, **83**, 2016 (1961).

(13) We are indebted to Dr. J. H. Shoolery of Varian Associates for assistance in interpreting this part of the spectrum.

(14) A particularly interesting feature of these spectra is the apparent low order (0 and 0.2 c.p.s.), of coupling between the proton on C-2 and those on C-3.

(15) By the reaction of I-A with brosyl chloride in $CHCl_3$ in the presence of ethyldiisopropylamine.

(16) A. Tulinsky, *J. Am. Chem. Soc.*, **84**, 3188 (1962).

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RECEIVED JUNE 27, 1962

THE STRUCTURE OF MITOMYCIN A[1]

Sir:

The preceding communication reports the elucidation of the gross structure of the mitomycins by chemical means. This one represents a preliminary account of the X-ray structure determination of the N-brosyl derivative of mitomycin A, $C_{22}H_{22}N_3O_8SBr$. When the determination was initiated (Oct. 13, 1961), the author knew only that the molecule was some complex fused ring system with the brosyl group linked to nitrogen.[2]

Slow evaporation from a solution in methylene chloride and benzene (1:4) yielded monoclinic crystals. The space group eventually was fixed to be C2, a = 19.70 Å., b = 8.24 Å., c = 16.05 A., β = 95.80°, with 4 molecules/unit cell; empirical formula, $C_{25}H_{25}N_3O_8SBr$. The carbon content (high by three) and density measurements (X-ray molecular weight, 605.6) indicated solvation with half a mole of benzene.[2] Intensity data were collected using the stationary crystal-stationary counter technique employing balanced filters; of the 1,481 reflections accessible to 1 A. resolution, 1,352 (91.3%) were taken to be observable.

The structure was solved by the heavy atom method. To enhance the heavy atom vectors, the Patterson coefficients were sharpened with $(z_{Br}/f_{Br})^2$, where z_{Br} and f_{Br} are, respectively, the atomic number and scattering form of bromine. The coördinates of the bromine and sulfur atoms were determined readily and the positions of the two carbons linked to them inferred. Phases based on these four atoms were employed to compute a 3-dimensional electron density (ρ_1). Two days afterward, (Nov. 17, 1961), this density was analyzed to contain 45 peaks greater than 1.7 e.A.$^{-3}$ (included carbons are in this count). Of these peaks, 30 belonged to the molecule, 7 were eliminated because of close approaches to either the brosyl system or the bromine atom; 2 others approached too close to a 2-fold rotation

(1) This research has been supported by the National Institutes of Health, U. S. Public Health Service and Lederle Laboratories Division of the American Cyanamid Company.

(2) The author wishes to thank the personnel of the Organic Chemical Research Section, Lederle Laboratories Division, American Cyanamid Company, for supplying samples and information, and especially Dr. J. H. Mowat, who furnished 8 mg. of this derivative and the crystallization method; and Mr. A. Mistretta, who demonstrated benzene of solvation by gas chromatography.