RADIOLYSIS OF METHANOL BY RECOILS FROM THE $B^{10}(n,\alpha)Li^7$ REACTION[1]

Sir:

The radiolysis of methanol has been the subject of much recent work.[2] Only one[2b] of these studies employed alpha particles. McDonell and Newton's work utilized 28 mev. cyclotron alphas, involved destruction of about 2% of the substrate and was carried out before the difficulty in achieving reproducibility in this system was recognized.

The work reported herein employed the alpha and Li^7 recoils of total energy 2.35 mev. per nuclear event resulting from the absorption of thermal neutrons by boron present in the form of dissolved methyl borate. Liquid methanol which was approximately 0.2 *M* in methyl borate was irradiated in the thermal-neutron facility[3] of the Brookhaven National Laboratory. Total energy absorbed from alpha particles was determined in two ways. In Method I, dosimeters consisting of aerated 0.001 *M* $FeSO_4$ in 0.8 *N* aqueous sulfuric acid containing 0.149 *M* boric acid were used in conjunction with the value of $G(Fe^{+3}) = 4.22$ determined by Schuler and Barr[4] for boron recoils in this system. Gamma background was determined simultaneously with boron-free Fricke dosimeters taking $G(Fe^{+3}) = 15.6$. It was assumed that the entire alpha plus lithium flux is absorbed in both the dosimeter solution and in methanol. No correction was applied for the gamma energy associated with alpha emission. Method II employed a scintillation counter calibrated by β-γ-coincidence counting of simultaneously neutron-irradiated gold foils.[4,5] After correction for attenuation of the neutron flux by boron,[4] the latter procedure yielded values that exceeded by about 7% those provided by Method I. The results of Method I were employed in calculating the *G*-values presented in Table I.

The methanol was Fisher's "Certified Reagent" which was purified, degassed and charged into quartz irradiation cells in essentially the same fashion as has already been described.[2h] Trimethyl borate (Metal Hydrides Co.) was purified by distillation using a 40 cm. column packed with glass helices. In the runs which provided the data of Table I, its concentration fell in the range 0.170 to 0.268*M*. Analytical procedures were similar to those that have already been described.[2h] Preliminary work[6] established that borate does not interfere with determination of ethylene glycol with the aid of chromotropic acid,[2h] but does interfere with determination of formaldehyde unless an increased amount of chromotropic acid (1 ml. of 25% solution) is employed.

The Saunders–Taylor micromanometric analysis was improved by use of CuO containing 1.3% of Fe_2O_3 which was prepared according to the method of Brückner and Schick[7] and was validated by analysis of known mixtures. Yields obtained in Co^{60} gamma radiolysis of methanol containing approx. 0.2 *M* methyl borate are also presented in Table I and were used in correcting for radiolysis by the gamma background present in the thermal column. Total recoil doses were about 3×10^{19} ev./ml.; gamma energy absorbed was about 7×10^{17} ev./ml. About 0.02% of the methanol was decomposed in the recoil radiolyses.

The data of Table I must be considered with caution in view of the uncertainty which still exists as to $G(H_2)$ and $G(CH_4)$ for Co^{60} gamma radiolysis of "pure" methanol. Borate appears to have little effect on $G(CH_4)$, $G(CO)$ and $G(C_2H_6O_2)$ of gamma radiolysis but appears to increase $G(CH_2O)$ significantly and probably reduces $G(H_2)$. The much greater LET of the recoil radiolysis as compared to gamma radiolysis has these consequences:

(1) $G(-CH_3OH)$ is somewhat *increased*
(2) $G(CO)$ increases at least tenfold and $G(CH_4)$ about twofold
(3) $G(C_2H_6O_2)$ is halved and the loss is only partially compensated by increase in $G(CH_2O)$

In the presence of 0.2 *M* borate, $G(Ox)$ exceeds $G(Red)$ by about 1 for both gamma ray and recoil particle radiolyses. The formation of an as yet unidentified reduction product or products is thereby suggested.

TABLE I

Energy source	Product yields, molecules/100 ev. H_2	CO	CH_4	CH_2O	$(CH_2OH)_2$
2.35 Mev. recoils[a]	5.14 ± 0.02	0.92 ± 0.06	0.67 ± 0.04	3.45 ± 0.10	1.40 ± 0.10
Co^{60} gamma[b]	4.40 ± 0.01	0.07 ± 0.01	0.31 ± 0.06	2.86 ± 0.17	2.85 ± 0.08

[a] Uncertainties are av. deviations from the mean of results of four radiolyses. [b] Two radiolyses in the presence of *ca.* 0.2 *M* methyl borate.

(1) Research carried out under the auspices of the U. S. Atomic Energy Commission, in part under Contract AT (30-1) 2383.

(2) (a) W. J. Skraba, J. C. Burr, Jr., and D. N. Hess, *J. Chem. Phys.*, **21**, 1296 (1953); (b) W. R. McDonell and A. S. Newton, THIS JOURNAL, **76**, 4651 (1954); (c) W. R. McDonell and S. Gordon, *J. Chem. Phys.*, **23**, 208 (1955); (d) W. R. McDonell, *ibid.*, **23**, 208 (1955); (e) G. Meshitsuka, K. Ouchi, K. Hirota and G. Kusumoto, *J. Chem. Soc. Japan*, **78**, 129 (1957); (f) G. Meshitsuka and M. Burton, *Radiation Research*, **8**, 285 (1958); (g) G. E. Adams and J. H. Baxendale, THIS JOURNAL, **80**, 4125 (1958); (h) N. N. Lichtin, *J. Phys. Chem.*, **63**, 1449 (1959).

(3) H. J. Curtis, S. R. Person, F. B. Oleson, J. E. Henkel and N. Delihas, *Nucleonics*, **14**, No. 2, 26 (1956).

(4) R. H. Schuler and N. F. Barr, THIS JOURNAL, **78**, 5756 (1956).

(5) H. H. Seliger and A. Schwebel, *Nucleonics*, **12**, No. 7, 54 (1954).

(6) Carried out by Miss B. Dudek under the sponsorship of a grant of the National Science Foundation's undergraduate research participation program.

(7) H. Brückner and R. Schick, *Gas u. Wasserfach*, **82**, 189 (1939).

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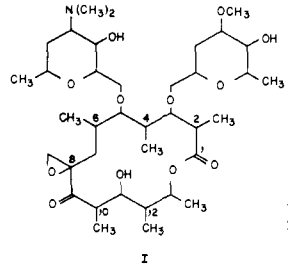
THE STRUCTURE OF OLEANDOMYCIN

Sir:

Oleandomycin, I, $C_{35}H_{61}NO_{12}$, is a macrolide antibiotic[1] which has been shown to contain the deoxysugars desosamine and L-oleandrose, glycosidically linked to a lactonic aglycone which possesses hy-

(1) This antibiotic, in the form of its triacetate ester, is known by the name Tao, a registered trade mark of Chas. Pfizer and Co., Inc.

droxyl, ketone and epoxide functions, and multiple C-methyl groups.[2]



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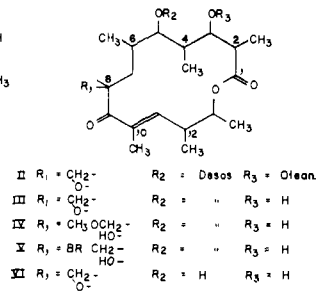


II R_1 = CH_2 (O), R_2 = Desos, R_3 = Olean.
III R_1 = CH_2 (O), R_2 = ", R_3 = H
IV R_1 = CH_3OCH_2, HO, R_2 = ", R_3 = H
V R_1 = BR CH_2, HO, R_2 = ", R_3 = H
VI R_1 = CH_2 (O), R_2 = H, R_3 = H

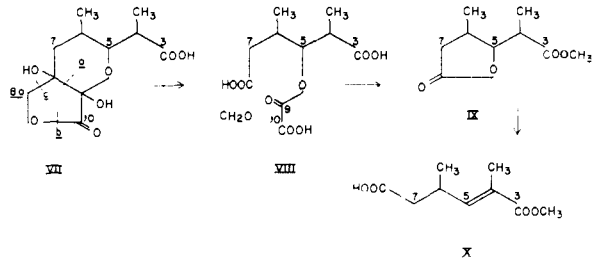
In mildly alkaline solutions the free hydroxyl group at C.11 of oleandomycin is lost by β-elimination to yield anhydroöleandomycin,[3] II, $C_{35}H_{59}NO_{11}$, m.p. 201–202°, λ_{max} 235 mμ, ϵ = 10,000. Hydrolysis of anhydroöleandomycin in methanol–sulfuric acid removes L-oleandrose to yield damylanhydrolide III, $C_{28}H_{47}NO_8$, m.p. 115°, and damylanhydrolidemethoxyhydrin, IV, $C_{29}H_{51}NO_9$, m.p. 79–80°, which has added methanol across the epoxide ring.

Damylanhydrolidebromohydrin, V, $C_{28}H_{48}NO_8Br\cdot HBr$, m.p. 149°, which results from the reaction of hydrobromic acid with damylanhydrolide is further hydrolyzed in benzene–aqueous hydrobromic acid yielding desosamine and anhydrolidebromohydrin, $C_{20}H_{33}O_6Br$, m.p. 166–167°. Anhydrolidebromohydrin is converted by base to the epoxide, anhydrolide, VI, $C_{20}H_{32}O_6$, m.p. 228–230°, λ_{max} 235 mμ, ϵ = 10,300, which, like anhydroöleandomycin is disubstituted α,β-unsaturated ketone.[4]

Extensive n.m.r. studies on anhydrolide derivatives and on appropriate model compounds established the presence of these several groups: (a) An exocyclic methylene epoxide converted by hydrogen chloride or hydrogen bromide to a primary alkyl halide (C.8, 8a); (b) two secondary hydroxyl groups (C.3, C.5); (c) a sequence X—CO—CMe=CH—CHX_2[5] (C.9, 10, 11, 12); (d) a sequence X_2CH—CHMe—O—COX (C.12, 13,1); (e) four C-methyl groups of type X_2CHMe, in addition to the C-methyls in *c* and *d*.

Nuclear magnetic resonance and ultraviolet spectra show that reduction of anhydrolide over platinum in acetic acid, or with sodium borohydride reduces the carbonyl group, but not the ethylenic double bond, to yield a dihydroisoanhydrolide, $C_{20}H_{34}O_6$, m.p. 165–167°. This dihydroisohydroanhydrolide yields no iodoform with hypoiodite, while dihydroisohydroanhydrolide acid, $C_{20}H_{36}O_7$, m.p. 173–174°, formed on hydrolysis, gives iodoform in good yield. This establishes the lactone oxygen and a C-methyl at C.13.

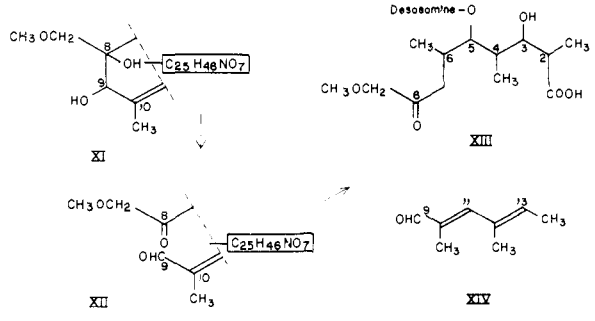
Periodate–permanganate oxidation[6] of damylanhydrolide, III, appears to oxygenate but not to cleave the C.10–11 double bond; nitric acid oxidation of the crude product hydrolyzes the epoxide ring and yields a new acid lactone, $C_{11}H_{16}O_7$, VII, m.p. 252–254°. This substance is oxidized by two equivalents of periodate at *a* and *c*, with hydrolysis at *b* to yield formaldehyde (C.8a) and the oily oxalate ester VIII. On successive base, acid and



VII VIII IX X

diazomethane treatment, VIII is hydrolyzed, lactonized and esterified to IX. Methoxide[7] opens IX to the 2,4-dimethyl-2-hexenedioic acid ester, X, m.p. 135–136.5°, whose structure was confirmed by synthesis. The acid lactone VII thus establishes the relationship of the C.8-8a epoxide, C.9 carbonyl, C.5 hydroxyl groups, and the C-methyls at C.4 and C.6. Exhaustive n.m.r. analysis of the methyl ester methyl ether, $C_{13}H_{20}O_7$, m.p. 173–175° formed by the reaction of VII with diazomethane confirms the structure of VII in detail, particularly the sequence X_3C—CH_2—CHX_2 (C.8,7,6). Nitric acid oxidation of oleandomycin itself yields, *inter alia*, (−)-methylsuccinic acid (C.5,6,7,8) and *meso*-3-hydroxy-2,4-dimethylglutaric acid (C.1,2,-3,4,5),[8] thus confirming the relationship of C.1 → C.10.

Hydrogenation and monoacetylation of IV yields dihydrodamylanhydrolidemethoxyhydrin monoacetate, XI, $C_{31}H_{55}NO_{10}\cdot CHCl_3$, pK_a = 6.9, m.p. 194–196°, which has a reduced carbonyl group, and acetylated desosamine.[9]



XI XII XIII XIV

XI is oxidized by one equivalent of periodate to a crude α,β-unsaturated aldehyde, XII, λ_{max} 227 mμ, ϵ = 19,000. In base, XII is deacetylated and hydrolyzed to the acid XIII, $C_{21}H_{39}NO_8\cdot H_2O$, m.p. 109–112°, together with the $\alpha,\beta,\gamma,\delta$-unsaturated al-

(2) Hans Els, W. D. Celmer and K. Murai, THIS JOURNAL, **80**, 3777 (1958).

(3) All compounds other than those designated as crude show satisfactory analyses and consistent ultraviolet and infrared spectra. All numbers in figures refer to the original positions in oleandomycin. Structures other than that of L-oleandrose are without stereochemical significance.

(4) R. B. Woodward, THIS JOURNAL, **63**, 1123 (1941); **64**, 72, 76 (1942).

(5) X is an unspecified group other than hydrogen.

(6) R. U. Lemieux and E. von Rudloff, *Can. J. Chem.*, **33**, 1701 (1955).

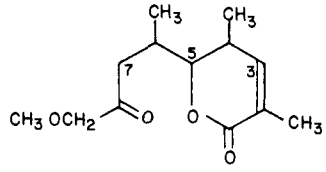
(7) See J. A. Elvidge and R. P. Linstead, *et al.*, *J. Chem. Soc.*, 2228 (1950), 3386 (1951); 1026 (1952), for analogous lactone cleavages.

(8) K. Gerzon, E. H. Flynn, M. V. Sigal, Jr., P. F. Wiley, R. Monohan and U. C. Quarck, THIS JOURNAL, **78**, 6396 (1956).

(9) Acid constants quoted by W. D. Celmer, "Antibiotics Annual," 1958–1959 Medical Encyclopedia, Inc., New York, N. Y., p. 277 (1959), and by W. D. Celmer and F. A. Hochstein, Abstracts of 133rd A.C.S. meeting, San Francisco, California, p. 20-M, April, 1958, support the assignment of the acetyl group to desosamine.

dehyde XIV, formed by vinylogous β-elimination of the C.13 oxygen. XIV was isolated as its dinitrophenylhydrazone, $C_{14}H_{16}N_4O_4$, m.p. 177–178° λ_{max} 265, 301 and 389 mμ.[10] This aldehyde establishes the position of the original ketone at C.9 relative to the lactone hydroxyl at C.13.

The acid, XIII, which does *not* lactonize, loses desosamine (C.5) and water (C.2–C.3) on benzene–hydrochloric acid hydrolysis and now lactonizes to the C.1–C.5 lactone, XV, $C_{13}H_{20}O_4$, dinitrophenylhydrazone m.p. 194–196°. The positions of the C.3 and C.5 hydroxyls relative to the carboxyl and the presence of desosamine at C.5 in XIII, and



XV

hence in oleandomycin are thus established. Oleandrose cannot be linked to desosamine,[2] and must therefore be attached at C.3, the only alternative site.

The structure of oleandomycin therefore is I.

(10) F. Bohlmann, *Ber.*, **84**, 490 (1951), quotes data for analogous compounds.

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REDUCTION OF POLYPEPTIDE-COBALT(III) COMPLEXES[1]

Sir:

The proposal that electron conduction may be a function of proteins has been an intriguing one.[2] We report here results suggesting the occurrence of some form of electron transfer through peptides during an oxidation-reduction reaction involving polypeptide ligands.

For the reduction of carboxylatopentamminocobaltic ions by chromous ion, Taube has shown that if the carboxylato moiety allows the reductant to donate its electron at a point remote from the cobalt (*e.g.*, some conjugated unsaturated acids), the consequent decrease in repulsion of the two ions at the transition state results in an increase in reaction rate several hundredfold.[3] We have applied this sensitive system to the polypeptide case, where a conduction process would reduce coulombic effects and provide a choice of sites at which an electron might be injected for reduction of a cobalt atom.

Two water-soluble copolymers of DL-alanine and L-glutamic acid, with monomer ratios 5.2 ala/glu and 2.7 ala/glu, were treated with aquopentamminocobaltic perchlorate. Dialysis afforded purified carboxylatopentamminocobaltic complexes, which exhibited absorption maxima identical with those of non-polymeric complexes such as acetatopentamminocobaltic ion (λ_{max} = 503 mμ, ϵ = 71).[4] No cobalt is bound by poly-DL-alanine itself under these conditions. The glutamic acid residues, and thus the cobalt atoms, are approximately randomly distributed, since the carboxyls of the 5.2/1 copolymer titrate in 0.1 *M* potassium chloride as independent groups with pK 4.0.

The cobalt-containing products, and their precursors, have a helical α-structure similar to that of poly-DL-alanine.[5,6] Films of these polymers undergo the infrared spectral changes shown by the homopolymer:[5] $\nu_{C=O}$ 1655 cm.$^{-1}$ (α form), converted to 1655, 1625 cm.$^{-1}$ (α + β) by water at 100°, back to 1655 cm.$^{-1}$ by formic acid. In deuterium oxide the polymers exhibit $\nu_{C=O}$ 1640 cm.$^{-1}$ (α) plus a shoulder at 1660 cm.$^{-1}$ corresponding to a solvated form.[7]

Reduction by chromous ion of these polymeric complexes and of several simpler analogs was followed by disappearance of the 500 mμ absorption. The results are given in Table I.

TABLE I

REDUCTION OF CARBOXYLATOPENTAMMINOCOBALTIC PERCHLORATES BY CHROMOUS ION[a]

$$Cr^{++} + [L\text{-}Co(NH_3)_5]^{++} \xrightarrow{k} [Cr\text{-}L]^{++} + Co^{++} + 5NH_3$$

L	T, °C.	k, M^{-1} sec.$^{-1}$	% of reaction
Acetate	25	0.15[b]	.....
Glycinate	26	.06	.....
Acetylglycinate	26	.30	.....
γ-Acetylaminobutyrate	26	.23	.....
Succinate	25	.19[b]	.....
Copolymer 5.2/1[c]	16.5	>40[e]	44–64[f]
		0.08	20[f]
Copolymer 2.7/1[d]	16.5	>40[e]	50–75[f]
		0.1	50–20[f]

[a] 0.5 *M* $HClO_4$, μ = 1.0, [Co] = 0.002–0.005 *M*, [Cr] = 0.005–0.02 *M*. Complete solution at all times. [b] Ref. 3. [c] Equivalent weight/Co, 560. [d] Equivalent weight/Co, 900. [e] Portion of reaction complete in less than 5 sec. [f] The relative fractions of fast reaction, transition period, and measurable second order rate varied from run to run. The smaller constants correspond to the last portion of reaction.

From the table, it will be seen that the first 50% of the reduction of the polymeric complexes occurs at least two hundred times faster than that of the monomeric complexes. One explanation of this rapid reaction may be that the transition state is stabilized by chelation of chromium. Against this interpretation may be argued the acidity of the medium and the evidence that systems in which chelation is possible, *e.g.*, the acetylglycine and suc-

(1) Supported by U. S. Public Health Service Grant E-1824 and National Science Foundation Grant 5717.

(2) A recent discussion of the subject appears in *Discussions Faraday Soc.*, **27**, (1959), "Energy Transfer with Special Reference to Biological Systems," Part II, p. 111 ff.

(3) H. Taube, "Mechanisms of Redox Reactions of Simple Chemistry," in "Advances in Inorganic and Radiochemistry," Vol. I, H. J. Emeleus and A. G. Sharpe, editors, Academic Press, Inc., New York, 1959, p. 25; H. Taube, *Can. J. Chem.*, **37**, 129 (1959).

(4) D. K. Sebera, Ph.D. Thesis, University of Chicago, 1959.

(5) A. Elliott, *Nature*, **170**, 1066 (1952).

(6) A. Berger and K. Linderstrøm-Lang, *Arch. Biochem. Biophys.*, **69**, 106 (1957).

(7) R. R. Blout and M. Idelson, THIS JOURNAL, **80**, 4909 (1959).