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THE STRUCTURE OF LUCENSOMYCIN. PART I

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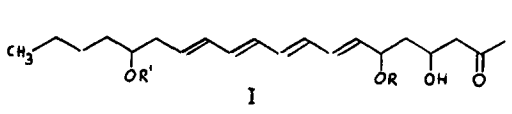
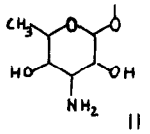
In a preliminary publication (1) we reported on the partial structure of lucensomycin, a tetraenoid macrocyclic antibiotic isolated from submerged cultures of Streptomyces lucensis. On the basis of the experiments summarized below, the following structural fragments were shown to be present in the antibiotic :



I



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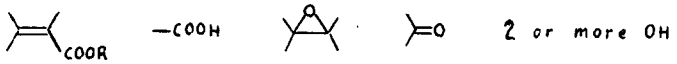
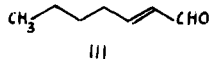
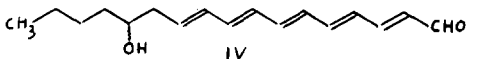


Ozonolysis of lucensomycin gave 2-heptenal (III), and alkaline treatment under mild conditions afforded 13-hydroxy-2,4,6,8,10-heptadecapentaenal (IV), indicating the presence in the antibiotic of an actual or potential grouping of type (I).



III



IV

The aldehyde (IV) could in fact arise from (I) by a retroaldolization process followed by β-elimination of OR. The presence of the residue (II) accounted for the formation of two acetates (tri- and tetra-) of mycosamine on acetlysis of lucensomycin with acetic anhydride and sulfuric acid. In addition to the occurrence of a carboxylate

anion band at 1585 cm^{-1} in the IR spectrum, the presence of a free carboxyl group was demonstrated by the amphoteric character of the antibiotic and by its easy decarboxylation in an acidic medium. Titration of the acylatable groups gave values corresponding to the presence of about 5 hydroxyls (in addition to the NH_2 group).

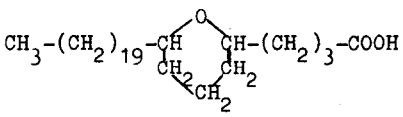
The presence of a ketonic function was inferred from the fact that lucensomycin did not show the retroaldolisation and decarboxylation reactions after treatment with $NaBH_4$. The presence of an α,β-unsaturated ester or lactone function was suggested by the occurrence in the UV spectrum of the antibiotic of a peak at 218 mμ, which disappeared on catalytic hydrogenation or on addition of thiols, but not on reduction with $NaBH_4$. Finally, the existence in lucensomycin of an epoxide ring adjacent to the carbonyl or to the α,β-unsaturated ester was rendered probable by the positive Bodforss reaction (2), which was not given by the hydrogenated antibiotic. Moreover dodecahydrolucensomycin, in which the ketonic group had not been affected by reduction, contained an extra hydroxyl group. On the basis of the chemical behaviour of the antibiotic, which proovsd to be very similar to that of pimaricin, we advanced the hypothesis that lucensomycin was a homologue of pimaricin. Analytical data, which suggested a formula $C_{36}H_{53}O_{14}N$ (or $C_{37}H_{55}O_{14}N$) containing 3-4 CH_2 groups more than pimaricin which, according to Ceder (3) possessed the formula $C_{33}H_{47}O_{14}N$, supported this view.

Subsequent investigations reported in this and in the following paper (4) have led us to the complete elucidation of the structure of lucensomycin.

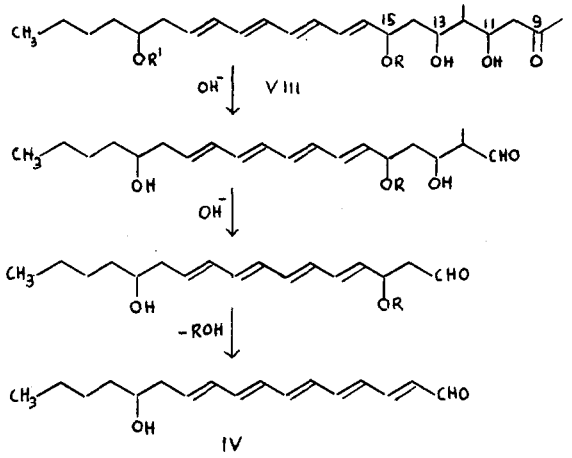
Hydrogenolysis of lucensomycin under drastic conditions, a technique developed by Ceder (3) (acetic acid solution at 250° and 200 atm, with Pt as the catalyst), affords n-nonacosanoic acid (V), 9-ketononacosanoic acid (VI) and 5,9-epoxynonacosanoic acid (VII) whose structures have been demonstrated by the mass and NMR spectra of the corresponding methyl esters.

$CH_3-(CH_2)_{27}-COOH$
V

$CH_3-(CH_2)_{19}-CO-(CH_2)_7-COOH$
VI


VII

The finding of (VI) among the hydrogenolysis products confirms the presence in lucensomycin of a ketonic function at carbon 9. That the carbonyl of the acid (VI) is the same as was originally present in the antibiotic is proved by the fact that hydrogenolysis of lucensomycin after treatment with $NaBH_4$ does not yield the acid (VI). Since the presence of a second ketonic function in the antibiotic is excluded, as will be shown later, the assignment of position 9 to the carbonyl leads to the conclusion that the 13-hydroxy-2,4,6,8,10-heptadecapentaenal (IV) results from a double retroaldolisation reaction. This implies the existence in lucensomycin of a system of type (VIII) from which the aldehyde (IV) is formed, as follows:



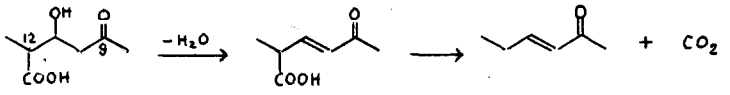
Moreover, the formation of the acid (VII) reveals the presence of an oxygenated function at carbon atom 5.

Reduction of dodecahydrolucensomycin with $LiAlH_4$ followed by treatment with phosphorus and iodine, and further reduction with $LiAlH_4$, affords n-nonacosane and 12-methylnonacosane (IX) :

$$CH_3-(CH_2)_{16}-\underset{\underset{CH_3}{|}}{CH}-(CH_2)_{10}-CH_3$$

IX

The production of this latter hydrocarbon demonstrates that a carboxyl function is attached at carbon 12 of the antibiotic. This carboxyl cannot be that of the α,β-unsaturated ester function contained in lucensomycin (maximum at 218 mμ in the UV spectrum) since, as shown by formula (VIII), carbon atoms 11 and 13 adjacent to carbon 12 both carry a hydroxyl. This fact locates the free carboxyl unequivocally at 12. On the other hand summation of the carbon atoms (30 contained in the hydrocarbon IX and 6 in mycosamine) and oxygen atoms of lucensomycin does not permit the presence of a second ester function in the antibiotic. The decarboxylation of the antibiotic on treatment with acids can be explained by the presence of a ketogroup at 9; this would promote the β-elimination of the hydroxyl in position 11 and the vinylogous β-ketoacid thus formed would undergo decarboxylation, as shown:



Lemieux oxidation (5) of lucensomycin (with $KMnO_4/KIO_4$) affords the oxalic acid hemiester (X) of 3-hydroxyheptanoic acid, which has been isolated in the form of its dimethyl ester :

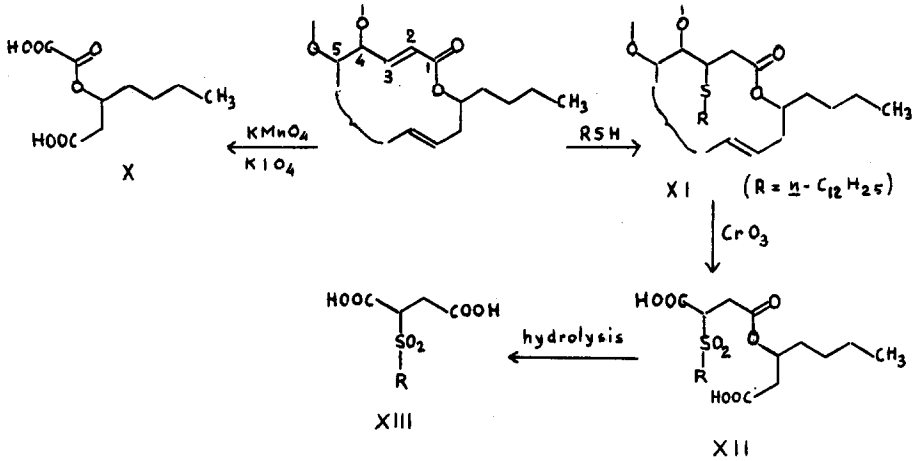
$$HOOC-CO-O-\underset{\underset{CH_2-COOH}{|}}{CH}-(CH_2)_3-CH_3$$

X

The structure of its ester, indicated by its NMR spectrum, has been demonstrated by comparison with an authentic synthetic sample (6). The formation of (X) provides direct evidence of the occurrence in lucensomycin of a lactonic function and locates the point of closure

of the lacton ring. Since no other ester function is present in the antibiotic, that which appears in (X) must be the same as that which on hydrogenolysis gives rise to the acids (V), (VI) and (VII); moreover, the 3-hydroxyheptanoic moiety of (X) represents the same fragment of the molecule of the antibiotic which is obtained in the form of 2-heptenal (III) by ozonolysis.

These conclusions are further supported by the fact that lucensomycin adds one mole of n-dodecylmercaptan to the double bond conjugated with the ester group (disappearance of the maximum at 218 mμ). Chromic acid oxidation of the sulphide (XI) thus obtained yields the sulphones (XII) and (XIII). All these reactions, which concern the same portion of the molecule of the antibiotic, are summarized below:

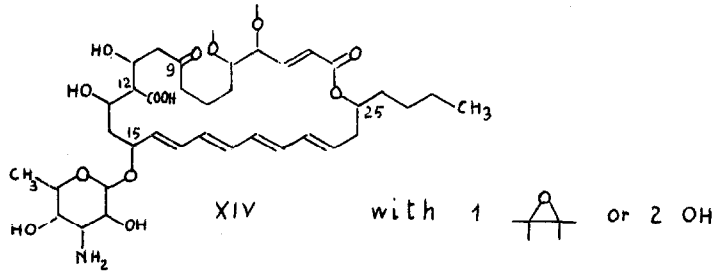


The structure of the sulphone (XII), isolated as its methyl ester, follows from its analysis and NMR spectrum. Chromic acid oxidation of (XI) gives, beside (XII), an appreciable quantity of n-dodecylsulphonylsuccinic acid (XIII), which clearly arises by hydrolysis of (XII). Among the oxidation products of (XI) we were unable to detect the presence, even in traces, of 3-n-dodecylsulphonylglutaric acid

which we had synthesized (6) for careful comparison. This fact would indicate the presence of an oxygen function at $C_{(14)}$, probably in the form of an epoxide, as previously postulated (positive Bodforss reaction). From the chromic acid oxidation of (XI), which contains, as previously demonstrated, an oxygen function at $C_{(5)}$, we might reasonably expect, beside (XIII), the formation of its homologue 3-n-dodecylsulphonylglutaric acid, unless an oxygen function is present on the adjacent carbon atom 4.

The demonstration that the site of closure of the lacton ring is carbon atom 25, together with the formation of 15-hydroxy-2,4,6,8,10-heptadecapentaenal (IV) by the alkaline treatment already discussed, leads to the conclusion that the mycosamine moiety is linked to the macrolide portion of the antibiotic at carbon atom 15.

On the basis of the experimental evidence and considerations above, we propose for lucensomycin the partial formula (XIV) :



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