NAPHTHOMYCIN, A NOVEL ANSA MACROCYCLIC ANTIMETABOLITE. PROTON NMR SPECTRA AND STRUCTURE ELUCIDATION USING LANTHANIDE SHIFT REAGENT

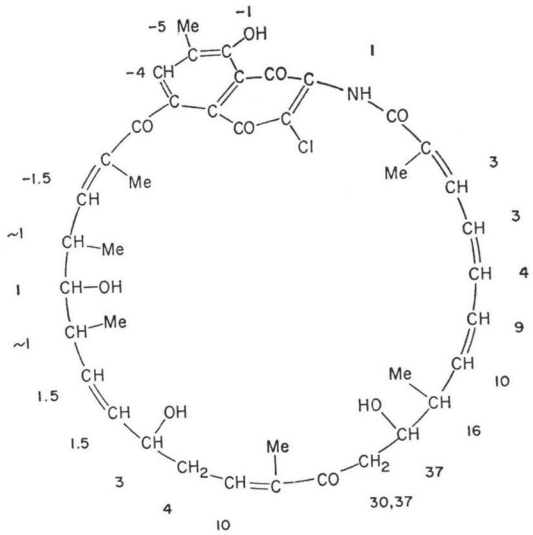
Sir:

The ansamycins* represent some of the most complex antibiotics.[1] They and their derivatives have aroused considerable interest as antiviral and antimicrobial agents, and as inhibitors of RNA tumor virus reverse transcriptases.[2] Hence, the potential significance of various structural features of these antibiotics and their derivatives in relationship to their biological activity is being investigated. We report herein the constitutional structure of naphthomycin[3] and show that it belongs to the class of naphthalenic ansamycins.

An antibiotic was isolated from the fermentation broth of an unidentified species of streptomyces. The compound showed antimicrobial activity against *Bacillus subtilis* in a chemically defined minimal medium and growth inhibition was relieved by addition of cysteine to the test medium. The biological and chemical properties showed a marked similarity with naphthomycin.[3] Chemical identity was confirmed by a comparison with an authentic sample kindly supplied by Dr. W. KELLER-SCHIERLEIN. The antibiotic used in our study was isolated from *Streptomyces* sp. X-12384 by solvent extraction and chromatographic methods in our Microbiology Department.

Naphthomycin, $C_{40}H_{46}ClNO_9$, $[\alpha]_{25}^{D}+432°$ (c 0.5, $CHCl_3$), dissolved in deuteromethylene chloride (50 mg in 0.45 ml), gave the following proton magnetic resonance data: δ TMS 0.94, 1.16, 0.80 (d, 3 each, J=6.5 Hz, $C\underline{H}_3CH<_{a,b,c}$ respectively), 2.25 (m, 2, H_a and H_b), 2.63 (m, 1, H_c), 2.01, 2.15, 1.71, 2.39 (s, 3 each, $C\underline{H}_3C=CH_{(4)}$, $C\underline{H}_3C=CH_{(8)}$, $C\underline{H}_3C=CH_{(9)}$, $C\underline{H}_3C=CH_{(10)}$, respectively), 2.30 (t, 2, $J_f=J_9=6.5$ Hz, $CH_f-C\underline{H}_2-CH_{(9)}=$), 2.53, 3.21 ($\underline{AB}H_e$, 2, J_{AB} *gem*=16.5, J_{Ae} *vic*=2.5, and J_{Be} *vic*=7 Hz, $CH_e-C\underline{H}_2-CO$), 2.80 (s, 1, OH), 3.75 (broad, 2, 2OH), 3.08 (dd, 1, J_{ad}=9.5 and J_{dc}=2 Hz, H'_d), 3.53 (ddd, 1, J *vic* to CH_2=2.5 and 7, and J_{be}=9.5 Hz, H_e), 3.98 (q, 1, $J_{CH_2}=J_{3f}=6.5$ Hz, H_f), 5.44, 5.59 (dAB, 2, $J_{1,3}$ *trans*=15, J_{1a}=8, J_{3f}=6.5 Hz, H_1 and H_3), 5.47 (dd, 1, $J_{2,7}$ *trans*=15 and J_{2b}=10 Hz, H_2), 5.95 (d, 1, J_{4c}=10 Hz, H_4), 6.12 ($\underline{AA'}XY$, 2, $J_{5,6}$ *cis*=12 Hz, H_5 and H_6), 6.53 (m, 1, $J_{2,7}$=15 Hz, H_7), 6.65 (m, 1, H_8), 6.76 (t, 1, J_{CH_2}=6.5 Hz, H_9), 7.98 (s, 1, aromatic H), 8.34 (s, 1, NH-CO), 9.63 (s, 1, OH). The above assignments were facilitated by spectral clarification induced by step-wise addition of 10 mg of a commercially available lanthanide shift reagent, *tris* (6, 6, 7, 7, 8, 8, 8-heptafluoro-2, 2-dimethyl-3, 5-octandionato) europium (III).[4]

Fig. 1. The constitutional structure of naphthomycin and relative induced shifts.



It resembles maytansine[2] which possesses a chlorine *ortho* to the amide nitrogen. Its structural features include an extended conjugated secondary amide function as in geldanamycin[5] and rifamycin S.[6] It possesses the 8-hydroxy-7-methyl-1, 4-naphthoquinone moiety of rifamycin S, but remarkably is not oxygenated at the 6-position. The carbon on C-5 bears an 8-carbon fragment with the same constitution as a group in streptovaricin D.[7] Finally, its bridge-spin of 23 skeletal carbon atoms exceeds dramatically not only 15 found for the benzenic maytansine[2] and

* The term "ansamycins" has been suggested by Prof. V. PRELOG for the new class of antibiotics containing an aliphatic ansa bridge, a bridge containing two non-adjacent positions of an aromatic nucleus.

geldanamycin,[5] but also 17 found for the naphthalenic rifamycins,[6] streptovaricins[7] and tolypomycins.[8]

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