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THE STRUCTURE OF ADENOMYCIN (C_{19-97} SUBSTANCE)

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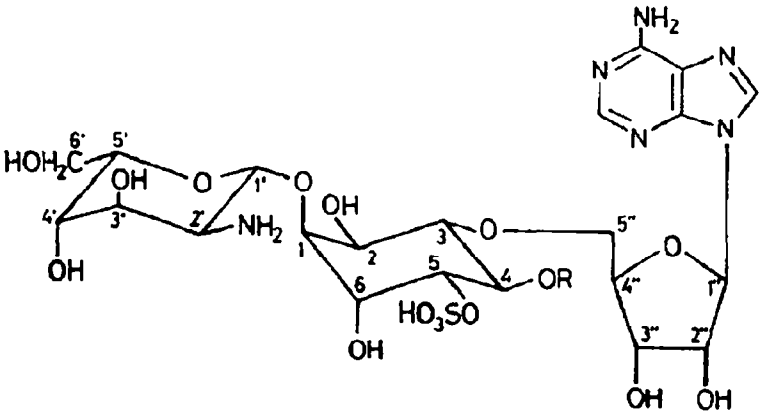
Summary. The structure of adenomycin, a new antibiotic active against Mycobacterium, has been established as a nuclecside consisting of adenine, D-ribose, (-)-chiro-inositol, L-gulosamine, L-serine and sulfate.

Adenomycin (1), an antibiotic active against Mycobacterium smegmatis is produced by Streptomyces griseoflavus.[1] This communication concerns the structural elucidation of 1.

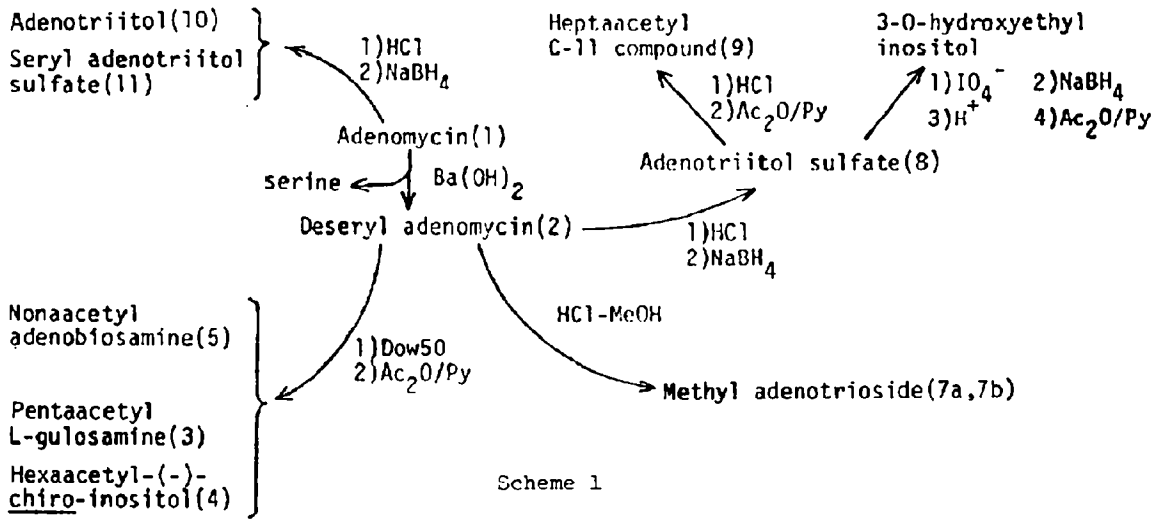
1, mp 165-168°C (dec.), $[\alpha]_D^{22.5}$ +10.5° (c 2, H_2O), $C_{25}H_{39}N_7O_{18}S$ (^{252}Cf plasma desorption MS[2] $(M-1)^-$ m/e 756), $\lambda_{max}^{H_2O}$ 260 nm (ε 11400), ν_{max}^{KBr} 3300 (OH and NH_2), 1750 (ester), 1690 (C=N), 1240 and 820 cm^{-1} ($O-SO_3H$);[3] was demonstrated to be an adenine nucleoside based on the spectral data of ^1H-nmr ($\delta_H^{D_2O}$ 8.40 and 8.22), ^{13}C-nmr and UV spectroscopy. The adenine nucleus was deduced to link to the sugar moiety through N-9 by comparison of the ^{13}C-nmr spectra of 1 with that of adenosine.[4] The summarized degradation studies are presented in Scheme 1.



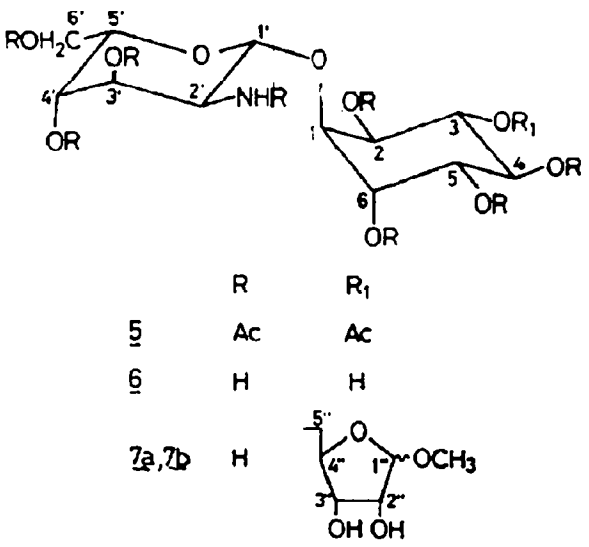
1 R: -C(=O)-CH(NH2)-CH2OH

2 R: H

Fig. 1



Scheme 1



	R	R_1
5	Ac	Ac
6	H	H
7a,7b	H	

Fig. 2

The ester cleavage of 1 by $Ba(OH)_2$ afforded serine and deseryladenomycin [(2), $C_{22}H_{34}N_6O_{16}S$, $\lambda_{max}^{H_2O}$ 260 nm (ε 1427($\nu_{max}^{H_2O}$ 3300, 1690, 1240 and 820 cm^{-1}] whose structure 2 was established as tha depicted in Fig. 1 based on the results described below.

Acid hydrolysis (Dowex 50W, H^+) of followed by acetylation (Ac_2O/Py) gave pentaacetyl L-gulosamine (3), hexaacetyl (-)-chiro-inositol (4) and nonaacetyl adenobiosamine (5), $C_{30}H_{41}NO_{19}$. The structure of 5 shown in Fig. 2 was elucidated by degradative studies and spectral evidence which showed the glycosidic linkage to be between C-1' o gulosamine [$\delta_C^{CDCl_3}$ 100.6, $\delta_H^{CDCl_3}$ 4.77 (d, J=9.8Hz)] and C-1 of inositol [δ_C^{CDC} 74.5, $\delta_H^{CDCl_3}$ 4.14(dd, J=3.0, 3.9Hz)]. In addition, the presence of a C-5 sugar was suggested by the molecular formula of 2 and adenobiosamine (6), $C_{12}H_{23}NO_{10}$.

Methanolysis of 2 gave methyl α- and β-adenotrioside (7a and 7b), $C_{18}H_{33}NO_{14}$. The presence of the adenobiosamine moiety plus the C-5 sugar in 7a and 7b was demonstrated by degradative studies as well as spectral evidence. The ^{13}C-nmr spectra of 7a and 7b showed four characteristic signals having a longer T_1 value. Since these signals showed close correspondence to the resonances of methyl (5-O-methyl) α and β ribofuranoside,[5] the C-5 sugar in 7a and 7b was assig to ribose. In addition, the signals of the alkylated oxymethylene at 72-74 ppm in the spectrum of 7a and 7b were assigned to C-5" of the ribose moiety. These results together with the mass

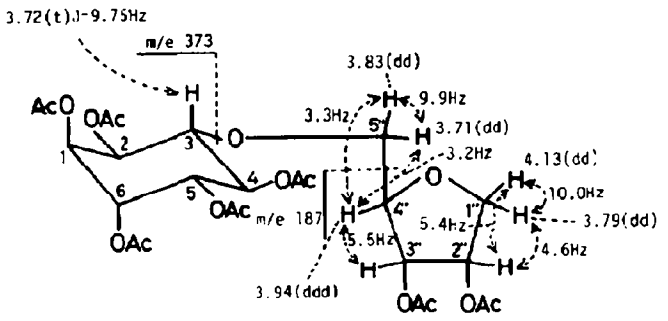


Fig. 3

spectral data of the peracetates of 7a and 7b indicate an ether linkage between the C-5" of ribose and one of the hydroxyl groups of inositol, leading to the structure of 7a and 7b (Fig. 2). The substitution site of C-5" to the inositol moiety was confirmed by the following experiments; acid hydrolysis of 2 followed by $NaBH_4$ reduction gave adenotriitcl sulfate (8), $C_{17}H_{33}NO_{17}S$, ν_{max}^{KBr} 3300, 1240, 820 cm^{-1}, which further gave heptaacetyl C-11 compcund (9), $C_{25}H_{34}O_{16}$, on acid hydrolysis followed by acetylation. The structure of 9 was elucidated to be a pseudodisaccharide consisting of inositol and 1,4-anhydroribitol on the basis of the ^{1}H-nmr, ^{13}C-nmr and mass spectral evidence. This is shown in Fig. 3 in which ribitol is linked to C3 (or C4) of inositol [$\delta_H^{CDCl_3}$ 3.72(t, J=9.75Hz)]. These results indicate that the ribose moiety is bonded to C3 or C4 of the inositol moiety in 1 and lead to the partial structure dipicted in Fig. 4.

The binding sites of ribose, sulfate and serine were determined by structural analysis of the three compounds which were obtained by acid hydrolysis of 1 and 2 followed by $NaBH_4$ reduction, viz, adenotriitol(10), $C_{17}H_{33}NO_{14}$, ν_{max}^{KBr} 3300 cm^{-1}, adenotriitol sulfate(8), $C_{17}H_{33}NO_{17}S$, ν_{max}^{KBr} 3300, 1240 and 820 cm^{-1}, and seryl adenotriitol sulfate(11), $C_{20}H_{38}N_2O_{19}S$, ν_{max}^{KBr} 3300, 1750, 1240 and 820 cm^{-1}.

The structure of 10 was established to be that dipicted in Fig. 5 by comparison of the ^{13}C-nmr spectra of 10 with those of 7a, 7b and 5-O-methyl ribitol. The structural similarities of 10, 8 and 11 were suggested by the ^{13}C-nmr of these compounds as shown in Fig. 5. The difference between 10 and 8 was demonstrated to be due to the presence of a sulfate in the latter by spectral evidence. Analogously, the difference between 8 and 11 was deduced to be due to the presence of serine in the latter.

Periodate oxidation of 8 followed by $NaBH_4$ reduction, acid hydrolysis and acetylation afforded hexaacetyl-3-O-hydroxyethyl inositol(12). Since this result indicates the absence of vic-diol in inositol, the substituents (L-gulosamine, ribitol and sulfate) must occupy C-1, C-3 and C-5 of the inositol. These results established the structure of 8 to be that shown in Fig. 5 and also confirmed the binding

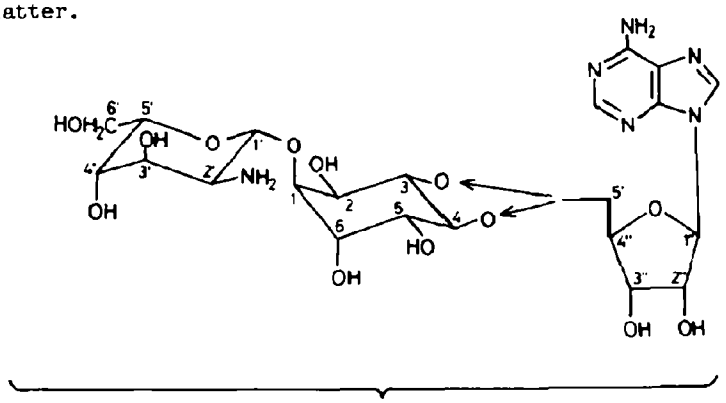


Fig. 4

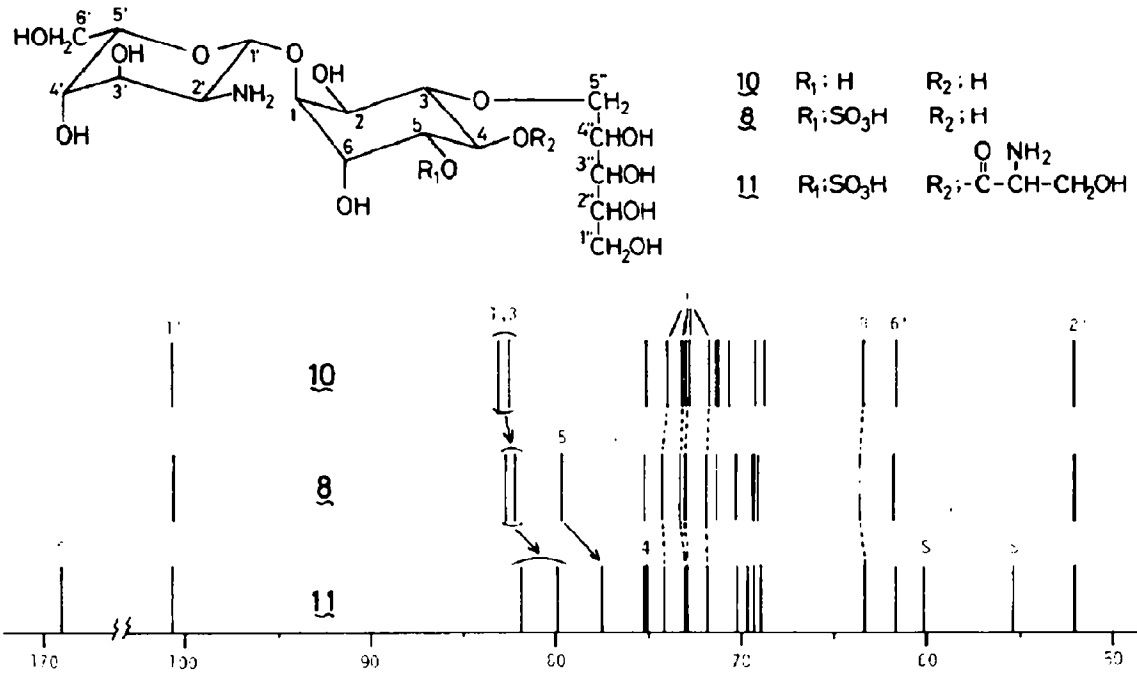


Fig. 5

site of ribose and sulfate in 1.

The structure of 11 was elucidated by comparison of the ^{13}C-nmr of 11 with that of 8. The signal assigned to C-5 shifts upfield in the spectrum of 11 was ascribed to be the γ-shift of acylation indicating that the substitution site of serine was at either C-4 or C-6 of insitol. Identification of the binding of serine at C-4 was achieved by inspection of the ^{1}H-nmr of 11, in which the characteristic signal (δ^{D_2O} 5.38, t, J=9.8Hz) due to the proton bearing serine coupled with both protons of C-3 and C-5 was observed. On the basis of this evidence, the structure of 11 as well as the binding site of serine was established.

The stereochemistries of serine and ribose were determined to be L and D, respectively, by ORD and CD spectra.

Based on all of these results, the structure of 1 was proposed as that depicted in Figure 1.

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