Studies on the Structure of Chromomycin A_3 by ^1H and ^{13}C Nuclear Magnetic Resonance Spectroscopy

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The applications of various ^1H and ^{13}C n.m.r. spectroscopic techniques including ^{13}C spin–lattice relaxation time studies has confirmed the gross structure of chromomycin A_3 but has led to a revision of the nature of the glycosidic linkage between two of the sugar entities.

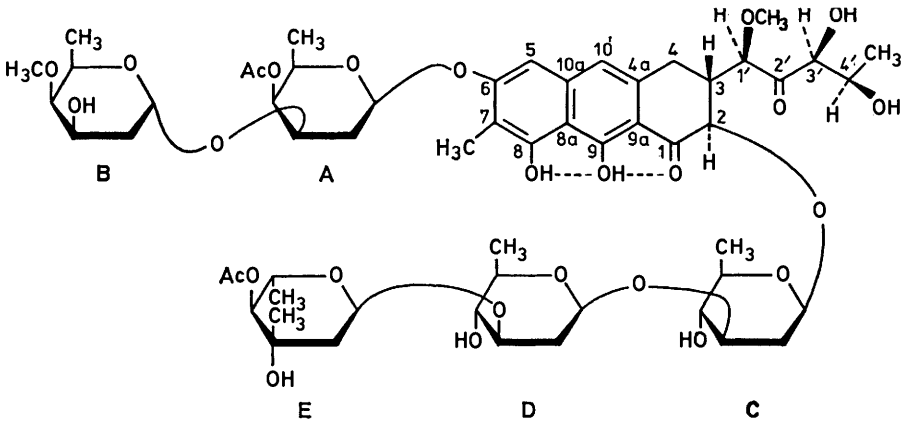
THE chromomycins,[1] olivomycins,[2] and mithramycin [3] represent a group of structurally closely related antibiotics showing marked antitumor properties. Their general structure consists of several different 2,6-dideoxysaccharides which form oligodeoxyoligosaccharide chains. These in turn are linked glycosidically to the aglycones chromomycinone or olivin (demethylchromomycinone), respectively. The structure investigation of chromomycin A_3 (1) by extended n.m.r. spectroscopy is the subject of the present study.

Whereas the structure of (1) is in general agreement with that originally proposed, based on hydrolyses and application of Klyne's rule,[1] an important revision concerns the interglycoside linkage of carbohydrate A (4-*O*-acetyl-2,6-dideoxy-D-*lyxo*-hexopyranose) and B (2,6-dideoxy-4-*O*-methyl-D-*lyxo*-hexopyranose). It could be demonstrated unequivocally that ring B is linked α-glycosidically to position 3 of carbohydrate A [as depicted in the disaccharide unit (2)], in contrast to the suggested [1] α,1 ⟶ 4-bond between carbohydrate B and 3-*O*-acetyl-2,6-dideoxy-D-*lyxo*-hexopyranose [as depicted in the disaccharide unit (3)].

^1H N.m.r. spectra of (1) in $CDCl_3$ or in CD_2Cl_2 (Figure 1) showed a nice separation of signals, whereas in solvents like [$^2H_6$]benzene, [$^2H_4$]methanol, or [$^2H_6$]acetone the full assignment of protons was not feasible. By detailed double resonance experiments the protons coupled to each other could be clearly assigned (*cf.* Table 1). In several instances superimposed signals were observed separately by running partially relaxed spectra.[4] Some of the axial 2-H atoms of the carbohydrate units were successfully assigned by application of the pulsed INDOR spectroscopy technique.[5]

In the aglycone a *trans*-diaxial arrangement of 2- and 3-H ($J_{2,3}$ 11.8 Hz) and a large coupling constant, $J_{3,4a}$ 12.3 Hz, were observed. These data were in agreement with a half-chair conformation of the condensed cyclohexenone ring and the formerly established [1] configurations at C-2*R* and C-3*S*. The observed allylic coupling constant $J_{4a,10}$ −1.5 Hz allows discrimination between 5- and 10-H.

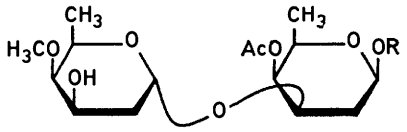
The assignment of the remaining functional groups such as the aromatic 7-methyl group as well as the hydrogen-bonded phenolic 8- and 9-OH groups was straightforward. However, an analysis of the protons in the complex side-chain with respect to its configuration appeared to be more difficult. The addition of one drop of [$^2H_4$]methanol turned out to be helpful resulting in considerable narrower line widths and distinguishable signals. Presumably without [$^2H_4$]methanol the hydrogen-bonded protons of the 3′- and 4′-OH groups undergo a rather slow exchange at 270 MHz and room temperature. A noticeable change in the chemical shifts of the protons in the side-chain upon addition of [$^2H_4$]methanol was not observed, and the coupling constants could be determined (*cf.* Table 1). The assignments of the 3′- and 4′-OH groups are arbitrary and may be interchanged. An unequivocal proof of the stereochemistry in the side-chain cannot be given by n.m.r. spectroscopy. However, the chemical shifts of the 3′- and 4′-OH groups strongly suggest hydrogen bonds to the C-2′ carbonyl group. With this assumption



(1)

the observed coupling constant, $J_{3',4'}$ 3.2 Hz, is in agreement with the formerly demonstrated [6] *threo*-configuration at C-3′ and -4′.

For the carbohydrate units in chromomycin A_3 (1) three β- and two α-anomers are readily observed as expected from the originally proposed structure.[1] The chemical shifts of the anomeric protons in two of the β-linked saccharides, δ(1A-H) 5.24 and δ(1C-H) 5.10,



(2)

(3)

clearly prove linkages of these carbohydrate units to C-2 or -6 of the aglycone, respectively. Their enhanced downfield shifts are attributed either to the C-1 carbonyl group or to the aromatic ring system. The carbohydrate hydroxy groups were not observed in the spectrum because of the fast exchange of all non-hydrogen-bonded OH groups due to the acidic phenolic functions. The neighbouring protons of the individual carbohydrate units were determined by extended double resonance experiments.

The coupling constants of 1A-H (carbohydrate A), $J_{1A,2aA}$ 8.5 and $J_{1A,2eA}$ 3.5 Hz, together with the considerable downfield shift, δ(1A-H) 5.24, prove the β-glycosidic linkage of saccharide A to the aglycone. The other coupling constants, $J_{2aA,3A}$ 11.3, $J_{3A,4A}$ 2.8, and $J_{4A,5A}$ 0.8 Hz, are in accord with a *lyxo*-configuration. The remarkably large downfield shift of 4A-H [δ(4A-H) 5.16] and the position of the 3A-H signal [δ(3A-H) 4.02] indicate the presence of a 4-acetoxy group. This is in contrast to the formerly suggested presence of a 3-acetoxy group, which was based on hydrolysis studies.[1]

In the carbohydrate unit B the coupling constants, $J_{1B,2aB}$ 2.7 and $J_{1B,2eB}$ 3.7 Hz, give evidence for an α-anomeric linkage. Again, the other coupling constants, $J_{2aB,3B}$ 11.0, $J_{3B,4B}$ 2.8, and $J_{4B,5B}$ 0.8 Hz, prove the *lyxo*-configuration for the saccharide B. The assignment of 1-H to the corresponding carbohydrate unit by double resonance experiments for carbohydrates A and B was supported by the large difference in the T_1 values of the 5-H atoms. In the α-linked saccharide B with an equatorial 1B-H the relaxation time for 5B-H (0.48 s) is approximately 50% longer than for 5A-H (0.33 s) of the β-linked saccharide A with an axial 1A-H. A straightforward calculation of relaxation times using a Dreiding model of (2) led to the same ratio of T_1(5A-H) : T_1(5B-H) as in the experiment. The relatively small difference in chemical shifts between 3A- and 3B-H (Δδ 0.13), as well as the large difference between 4A- and 4B-H (Δδ 1.96) support the presence of a 4-acetoxy group in sac-

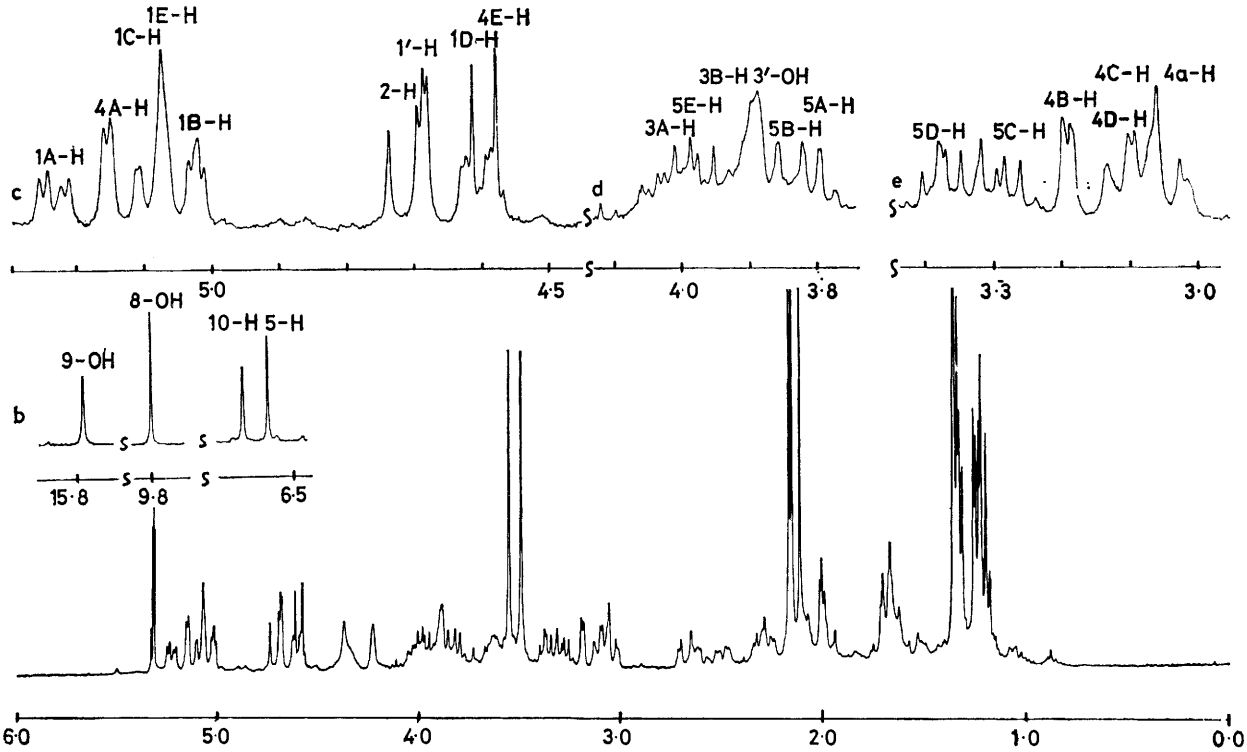


FIGURE 1 ^{1}H N.m.r. spectrum of chromomycin A_3 (1) in CD_2Cl_2 at 270 MHz; a, normal spectrum; b, downfield signals (normal spectrum); c—e, expanded proton regions

TABLE 1

^{1}H N.m.r. data of (1) [a]

Hydrogen atom	δ [b]	$J(H_i,H_j)$/Hz [c]	T_1/s [d]	Double resonance experiments [e]
Aglycone				
2-H	4.72 (d)	11.8 (2,3)	0.53 [f]	3
3-H	2.61 (dddd)	3.5 (3,4e), 1.8 (3,1′), 11.8 (3,2), 12.3 (3,2a)	0.27 [f]	(2,1′), 4a
4a-H [g]	3.07 (ddd)	−1.5 (4a, 10), 12.3 (4a, 3), −15.2 (4a, 4e)	0.19 [f]	(3, 4e), 10
4e-H	2.67 (dd)	3.5 (4e, 3), −15.2 (4e, 4a)	0.19 [f]	4a
5-H	6.55 (s)		0.48	
10-H	6.69 (d)	−1.5 (10, 4a)	0.56	4a
8-OH	9.75 (s)		0.74	
9-OH	15.75 (s)		1.02	
7-CH_3	2.17 (s)		0.54	
1′-H	4.69 (d)	1.8 (1′, 3)	0.43 [f]	(3, 4e)
3′-H	4.24 (d)	3.2 (3′, 4′)	0.43	
4′-H	4.35 (dq)	3.2 (4′, 3′), 6.2 (4′, 5′)	0.53	5′
5′-CH_3	1.34 (d)	6.2 (5′, 4′)	0.30	4′
3′-OH	3.89 (s)			
4′-OH	4.38 (s)			
Carbohydrate A				
1A-H	5.24 (dd)	8.5 (1, 2e), 3.5 (1, 2a)	0.31	(2a, 2e)
2aA-H 2eA-H	2.1 (m)			1
3A-H [g]	4.02 (ddd)	11.3 (3, 2a), 6.0 (3, 2e), 2.8 (3, 4)	0.43 [f]	(2a, 2e), 4
4A-H	5.16 (dd)	2.8 (4, 3), 0.8 (4, 5)	0.48	3, 5
5A-H	3.81 (dq)	0.8 (5, 4), 6.4 (5, 6)	0.33	4
6A-CH_3	1.24 (d)	6.4 (6, 5)	0.35	5
Carbohydrate B				
1B-H	5.03 (dd)	2.7 (1, 2a), 3.7 (1, 2e)	0.42	(2a, 2e)
2aB-H 2eB-H	2.0 (m)			1, 3
3B-H	3.89 (ddd)	2.8 (3, 4), 6.0 (3, 2e), 11.0 (3, 2a)		(2a, 2e), 4
4B-H	3.20 (dd)	2.8 (4, 3), 0.8 (4, 5)	0.48	(3, 5)
5B-H	3.87 (dq)	0.8 (5, 4), 6.6 (5, 6)	0.48	
6B-CH_3	1.25 (d)	6.6 (6, 5)	0.35	5
Carbohydrate C				
1C-H	5.10 (dd)	9.6 (1, 2a), 1.8 (1, 2e)	0.30	2a, 2e
2aC-H [h]	1.76 (ddd)	9.6 (2a, 1), −12.0 (2a, 2e), 12.0 (2a, 3)		1
2eC-H	2.50 (ddd)	1.6 (2e, 1), 5.0 (2 e, 3), −12.0 (2e, 2a)	0.22 [f]	2a, 3, (1C, 1E)
3C-H [g]	3.61 (ddd)	5.0 (3, 2e), 9.0 (3, 4), 12.0 (3, 2a)		2a, (4C, 4D, 4a)
4C-H	3.07 (dd)	9.0 (4, 3), 9.2 (4, 5)	0.58	3, 5
5C-H	3.30 (dq)	9.2 (5, 4), 6.2 (5, 6)	0.32	4, 6
6C-CH_3	1.33 (d)	6.2 (6, 5)	0.29	5
Carbohydrate D				
1D-H	4.61 (dd)	1.8 (1, 2e), 9.8 (1, 2a)	0.33 [f]	2a, 2e
2aD-H [h]	1.71 (ddd)	−12.0 (2a, 2e), 11.7 (2a, 3), 9.8 (2a, 1)		1, 2e, 3
2eD-H	2.28 (ddd)	1.8 (2e, 1), 5.2 (2e, 3), −12.0 (2e, 2a)	0.26 [f]	1, 2a, 3
3D-H [g]	3.53 (ddd)	5.2 (3, 2e), 9.0 (3, 4), 11.7 (3, 2a)		2a, 2e, (4D, 4C)
4D-H	3.11 (dd)	9.0 (4, 3), 9.0 (4, 5)	0.50	3
5D-H	3.38 (dq)	9.0 (5, 4), 6.3 (5, 6)	0.40	(4D, 4C), 6
6D-CH_3	1.35 (d)	6.3 (6, 5)	0.35	5
Carbohydrate E				
1E-H	5.08 (dd)	2.4 (1, 2a), 2.4 (1, 2e)		2a, 2e
2aE-H	1.67 (m)			(1E, 1C)
2eE-H	2.00 (m)			(1E, 1C)
4E-H	4.60 (d)	9.4 (4, 5)	0.72	5
5E-H	3.98 (dq)	9.4 (5, 4), 6.4 (5, 6)	0.66	(4E, 1D)
6E-CH_3	1.22 (d)	6.4 (6, 5)	0.38	5
3E-CH_3	1.36 (s)		0.36	
Functional groups				
1-OCH_3	3.56 (s)		0.69	
2-OCH_3	3.50 (s)		0.44	
1-$OCOCH_3$	2.16 (s)		0.69	
2-$OCOCH_3$	2.12 (s)		0.84	

[a] *c* 0.021 mol l^{-1} in CD_2Cl_2. 270 MHz. Total data memory 32 K, spectral width 4 424.8 Hz, *T* 296 K. [b] Chemical shifts downfield from Me_4Si (δ_{CDHCl_2} 5.32) and multiplicity of signals. [c] Coupling constants, coupled hydrogen atoms are given in parentheses. [d] T_1 Values are obtained from inversion recovery measurements by an exponential least-squares fit. [e] The numbers indicate the hydrogen atoms of the corresponding unit in which the irradiation is performed. If more than one signal is irradiated in one experiment, this is shown in parentheses. Hydrogen atoms of other units are denoted by head letters. [f] Calculated from zero-cross-over points. [g] Signal is obtained from a partially relaxed spectrum. [h] Signal is obtained from a pulsed INDOR experiment.

charide A. Thus, from these data it follows that the saccharides B and A in the disaccharide (2) are linked by an α,1 ⟶ 3-interglycoside bond.

As documented by the coupling constants (Table 1) the carbohydrates C and D show the described [1] β-*arabino* configurations. A considerable downfield shift is observed for the axial anomeric 1C-H of sugar C, probably caused by an anisotropy effect of the C-1 carbonyl group of the aglycone. The saccharide E is shown to be an α-anomer, $J_{1E,2^aE} = J_{1E,2^eE} = 2.4$ Hz, which carries an acetoxy group at C-4E [δ(4E-H) 4.60], and the appearance of 4E-H as a plain doublet gives evidence for branching at C-3E.

In conclusion the ^{1}H n.m.r. data are in agreement with the gross composition of chromomycin A_3 consisting of the aglycone chromomycinone, two molecules of 2,6-dideoxy-β-D-*arabino*-hexopyranose (carbohydrates C and D), one molecule of 4-*O*-acetyl-2,6-dideoxy-3-*C*-methyl-α-L-*arabino*-hexopyranose (carbohydrate E), one molecule of 2,6-dideoxy-4-*O*-methyl-α-D-*lyxo*-hexopyranose (carbohydrate B), and one molecule of 4-*O*-acetyl-2,6-dideoxy-β-D-*lyxo*-hexopyranose (carbohydrate A). The type of interglycoside linkages between the saccharide units cannot be deduced unequivocally by ^{1}H n.m.r. spectroscopy.

The signals from the ^{13}C n.m.r. spectrum were essentially assigned by selective double resonance experiments; and for non-hydrogen-bearing aromatic carbon atoms increment calculations were applied. The assignment of the carbohydrate deoxy C-2 atoms by selective double resonance experiments is prevented by higher order effects.

The ^{13}C n.m.r. spectrum of (1) and the assignments are depicted in Figure 2, and the parameters are nicely in accord with the results from the ^{1}H n.m.r. analysis. In the carbohydrate unit A the β-glycoside linkage is evident from the chemical shift of C-1A [δ(C-1A) 97.3] as well as from the coupling constant, $J_{C\text{-}1A,\,1A\text{-}H}$ 162.0 Hz.[7] Furthermore, the considerable downfield shift of C-3A [δ(C-3A) 82.3] by comparison with the chemical shift of C-3B [δ(C-3B) 65.8] proves that the glycoside bond of saccharide B is to C-3A of the saccharide A, which in turn leaves the acetoxy group at C-4A.

This could also be demonstrated by application of a selective double resonance technique,[8] which is depicted in Figure 3. Figure 3a shows an expansion of the carbonyl group region of both acetoxy groups at C-4A and -4E in the coupled ^{13}C n.m.r. spectrum. Both signals appear as a double quartet, the quartet splitting relating to the $^2J_{CCH}$ coupling with the acetyl methyl groups and the doublet to $^3J_{COCH}$ coupling with 4A- or 4E-H, respectively. By irradiation of 4A-H at low power the spectrum in Figure 3b is obtained. Obviously the doublet splitting is removed only in the acetyl carbonyl signal at δ 170.9, which gives evidence for an acetoxy group at C-4A in the saccharide A.

In the carbohydrate B the α-glycosidic bond is clearly deduced from the coupling constant, $J_{C\text{-}1B,1B\text{-}H}$ 167.0 Hz.

TABLE 2

^{13}C N.m.r. data of (1) [a]

Carbon atom	δ [b]	$^1J(^{13}C,H)$/Hz	$^nJ(^{13}C,H)$/Hz [c]	T_1/s [d]
Aglycone				
C-1	202.3 (s)			
C-2	76.1 (ddq)	148.0	3.5, 7.0	0.14 [e]
C-3	43.6 (ddd)	130.0	5.0, 9.0	
C-4	43.7 (tdd)	129.0	4.5, 7.5	
C-5	100.8 (ddd)	162.0	1.9, 5.0	0.28 [e]
C-6	159.5 (q)		4.0	
C-7	111.6 (qd)		6.0, 6.0	
C-8	164.2 (d)		3.0	
C-9	155.8 (t)		3.5	
C-10	117.2 (ddt)	161.0	3.0, 7.0	0.14 [e]
C-4a [f]	138.3 (s)			
C-8a [g]	108.0 (s)			
C-9a [g]	107.9 (s)			
C-10a [f]	134.6 (t)		6.0	
C-7l	8.1 (q)	128.0		
C-1′	79.5 (dt)	148.0	4.0	0.39
C-2′	211.3 (s)			
C-3′	78.2 (d)	148.0		0.22
C-4′	67.7 (d)	144.0		0.22
Carbohydrate A				
C-1A	97.3 (dddd)	162.0	4.0, 6.0, 9.0	0.25
C-3A	82.3 (d)	140.0		0.18
C-4A	67.3 (ddd)	148.0	5.0, 7.0	0.14 [e]
C-5A	69.7 (d)	144.0		0.19
$OCOCH_3$	170.9 (dq)		4.0, 7.0	
Carbohydrate B				
C-1B	96.9 (ddd)	167.0	2.0, 4.0	0.37
C-3B	65.8 (ddd)	144.0	5.0, 6.0	0.19
C-4B	81.5 (ddt)	140.0	4.0, 6.0	0.27
C-5B	69.9 (d)	140.0		0.17 [e]
Carbohydrate C				
C-1C	100.4 (dddd)	162.0	4.0, 8.5, 2.0	0.22
C-3C	82.0 (ddq)	140.0	4.0, 3.5	0.15 [e]
C-4C [h]	75.1 (d)	148.0		0.18 [e]
C-5C	72.3 (d)	141.0		0.15
Carbohydrate D				
C-1D	99.7 (ddd)	162.0	3.5, 7.5	0.23
C-3D	80.4 (d)	141.0		0.30
C-4D [h]	75.0 (d)	145.0		0.21
C-5D	72.1 (d)	144.0		0.22
Carbohydrate E				
C-1E	95.1 (dtd)	168.0	2.0, 4.0	0.29
C-3E	70.4 (dq)		3.5, 6.0	0.72 [e]
C-4E	67.0 (dt)	149.0	4.5	0.22
C-5E	66.8 (ddd)	145.0	7.0, 3.0	0.31
$OCOCH_3$	171.4 (dq)		3.5, 7.5	
Remaining atoms [i]				
CH_2	37.4 (t)	132.0		
CH_2	37.0 (t)	133.0		
CH_2	33.4 (tdd)	131.0	2.5, 4.0	
CH_2	32.9 (tdd)	133.0	2.0, 5.5	
CH_2	26.9 (tdd)	130.5	4.0, 7.0	
CH_3	23.0 (qdd)	129.0	5.0, 8.0	
CH_3	20.3 (q)	127.5		
CH_3	18.0 (q)	127.5		
CH_3	17.8 (q)	127.5		
CH_3	17.8 (q)	127.5		
CH_3	17.2 (q)	125.0		
CH_3	16.8 (qd)	127.5	2.0	
1-$OCOCH_3$	20.9 (q)	130.0		
2-$OCOCH_3$	20.7 (q)	130.0		
1-OCH_3	62.2 (qd)	142.0	7.0	0.64
2-OCH_3	59.6 (qd)	142.5	5.0	0.34

[a] *c* 0.056 mol l^{-1} in $CDCl_3$, 67.89 MHz. Total data memory 32 K, spectral width 16 129.0 Hz, *T* 302 K. [b] Chemical shifts downfield from Me_4Si (δ_{CDCl_3} 77.0) and multiplicity of signals. [c] In general long range coupling constants ($n \geqslant 2$) are not assigned to hydrogen atoms (see text). [d] T_1 Values are obtained from inversion recovery measurements using an exponential least-squares fit. [e] Calculated from zero-crossover points. [f–h] Assignments may be reversed. [i] Not assigned.

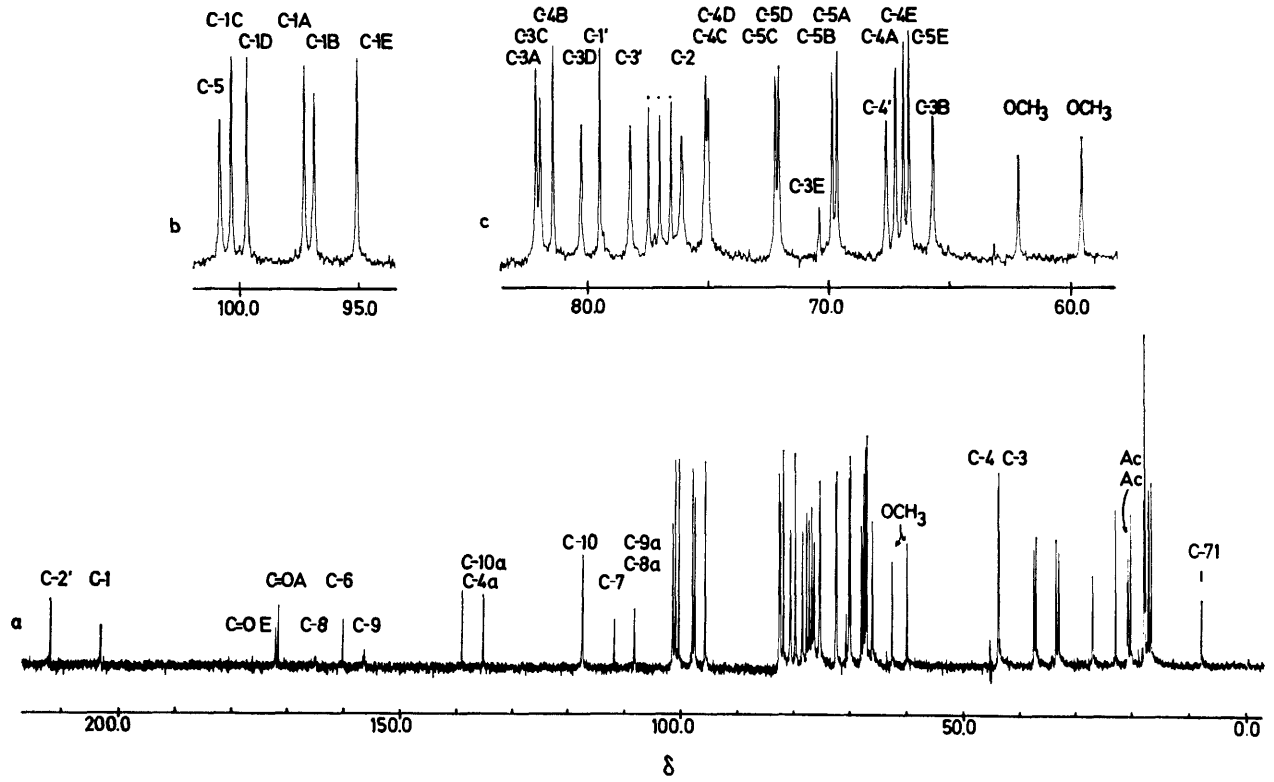


FIGURE 2 ^{13}C N.m.r. spectrum of chromomycin A_3 (1) in $CDCl_3$ at 67.89 MHz; a, noise decoupled spectrum; b, c, expanded carbon region

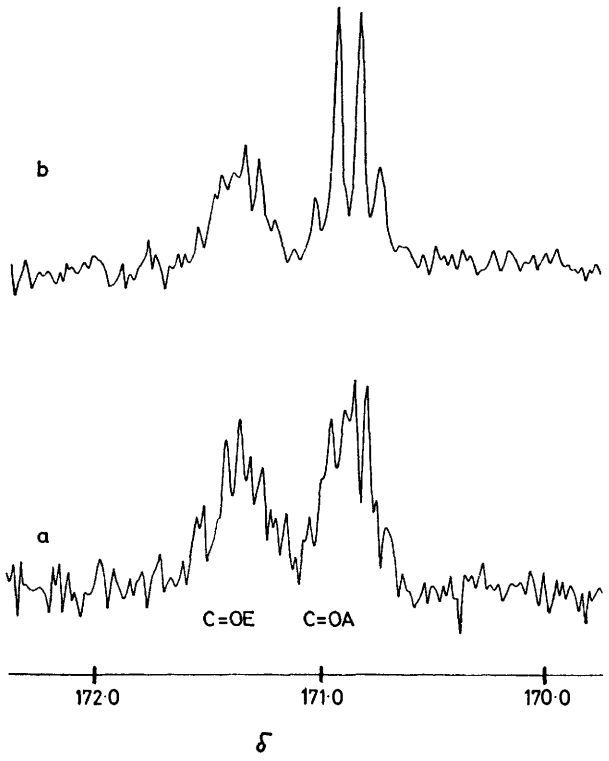


FIGURE 3 Selective double resonance experiment: a, expanded acetyl carbonyl region, gated-decoupled; b, as a plus irradiation of 4A-H (δ 5.17)

However, the difference in the chemical shifts between the anomeric carbon atoms in the α-linked saccharide B [δ(C-1B) 96.9] and the β-linked saccharide A [δ(C-1A) 97.3] turned out to be unusually low. The position of the methoxy group is unequivocally assigned by the chemical shift at C-4B [δ(C-4B) 81.5] which is in marked contrast to the chemical shift of the corresponding carbon of the other *lyxo*-compound A[δ(C-4A) 67.3].

By inspection of the ^{13}C n.m.r. data of the carbohydrates C and D their β-glycoside linkages are evident from the chemical shifts of the anomeric carbon atoms [δ(C-1C) 100.4 and δ(C-1D) 99.7] as well as from the corresponding coupling constants, $J_{C\text{-}1C,\ 1C\text{-}H} = J_{C\text{-}1D,\ 1D\text{-}H} = 162.0$ Hz. The chemical shifts of the C-3 atoms [δ(C-3C) 82.0 and δ(C-3D) 80.4] reveal that both saccharides C and D are further linked to other units *via* the 3-position.

In the saccharide E the α-glycoside linkage can be similarly assigned from the chemical shift of the anomeric carbon atom, δ(C-1E) 95.1, and the coupling constant, $J_{C\text{-}1E,\ 1E\text{-}H}$ 168.0 Hz. In addition to the above mentioned acetoxy group at C-4E the branching at C-3E is obvious from the singlet at δ(C-3E) 70.4 in the coupled spectrum.

From the ^{13}C n.m.r. spectra it can be deduced that the carbohydrates A, C, and D are inner saccharides, all of which are further linked to the other units *via* the 3-position. The carbohydrates B and E represent terminal

saccharides. Whereas for all the saccharides the functional groups and the type of anomeric linkage could be assigned, this method does not reveal information on their sequence. Several attempts for selective decoupling *via* the glycoside bonds [9] have not yet given unequivocal results.

Further ^{13}C spin–lattice relaxation time studies were performed in order to obtain information on the sugar sequence.[10] The average T_1 values ($\bar{T}_1$) of oxygen-bearing carbon atoms in the individual carbohydrate

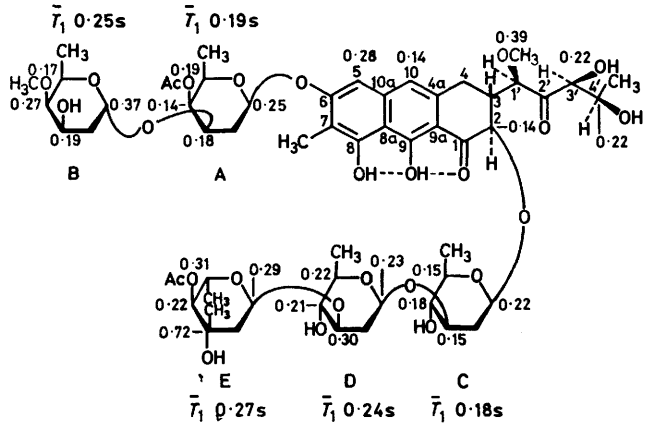


FIGURE 4 ^{13}C T_1 and average T_1 ($\bar{T}_1$) values of chromomycin A_3 (1). $\bar{T}_1$ of saccharide E is calculated by omitting T_1 (C–3E)

units were calculated (Figure 4). The shortest mean relaxation times are observed for the saccharides A ($\bar{T}_1$ 0.19 s) and C ($\bar{T}_1$ 0.18 s). This is fully in accord with the considerable downfield shift of 1A- and 1C-H caused by the aglycone moiety. Consequently the carbohydrates A and C represent the starting units of either saccharide chain.

As deduced from the ^{13}C chemical shifts the saccharides B and E were shown to be terminal saccharides. The saccharide D, however, turned out to represent an inner unit as is obvious from the ^{13}C and the ^1H n.m.r. data. Comparison of the average T_1 values of the saccharides B ($\bar{T}_1$ 0.25 s), D ($\bar{T}_1$ 0.24 s), and E ($\bar{T}_1$ 0.27 s) shows that the carbohydrate *E*, with the longest average T_1 value, occupies the terminal position of the trisaccharide chain. Furthermore, the saccharides B and D exhibit nearly the same mean relaxation times, which is in agreement with their secondary position in either saccharide chain. Subsequently the two saccharide chains in (1) consist of a disaccharide unit (2), α-B(1 → 3)-A, and a trisaccharide unit, α-E(1 → 3)-β-D(1 → 3)-C.

These results show chromomycin A_3 (1) to be (3*S*)-[(3*S*),(4R)-dihydroxy-(1*R*)-methoxy-2-oxopentyl]-3,4-dihydro-8,9-dihydroxy-7-methyl-1-oxo-2*H*-anthracen-(2*R*)-yl-[4-*O*-acetyl-2,6-dideoxy-3-*O*-(2,6-dideoxy-4-*O*-methyl-α-D-*lyxo*-hexopyranosyl)-β-D-*lyxo*-hexopyranosid]-6-yl-{2,6-dideoxy-3-*O*-[2,6-dideoxy-3-*O*-(4-*O*-acetyl-2,6-dideoxy-3-*C*-methyl-α-L-*arabino*-hexopyranosyl)-β-D-*arabino*-hexopyranosyl]-β-D-*arabino*-hexopyranoside}.

EXPERIMENTAL

^1H and ^{13}C n.m.r. spectra were recorded on a Bruker WH 270 spectrometer (^1H 270 MHz, ^{13}C 67.89 MHz). If not stated otherwise, generally a 90° measuring pulse was applied. The T_1 times were obtained by the inversion recovery technique using an exponential least-squares fit. The 90° pulses have a duration t_{13C} of 20 μs and t_{1H} of 10.8 μs. In pulsed INDOR experiments a selective 180° pulse of 0.15 s and a non-selective 40° pulse were applied. The delay between two 180° pulses was 4 s, between the selective and the non-selective pulses 0.01.

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