The Megalomicins. Part V.[1] Mass Spectral Studies

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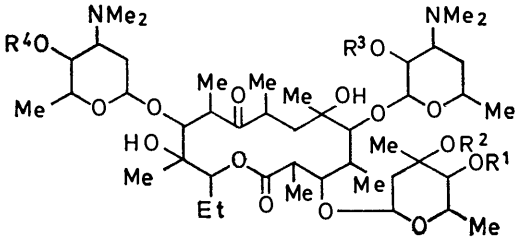
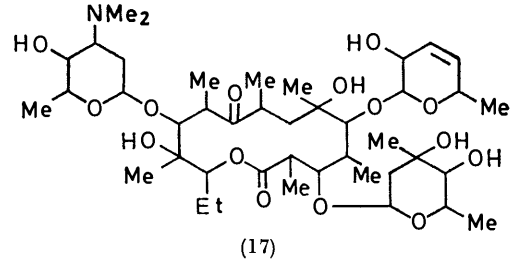
The mass spectral fragmentation patterns of the megalomicins, a new group of macrolide antibiotics elaborated by *Micromonospora megalomicea* sp. n., are described. The mass spectra of the 9-hydroxy-analogues, the megalalosamines, the erythralosamines, and erythromycin A are also discussed.

MASS SPECTROMETRY has been successfully used in the elucidation of the structures of a number of polyene antifungal antibiotics such as the amphotericins,[2] nystatin,[3] the mycoticins,[4] and the flavofungins,[5] and also for the macrolides neutramycin,[6] cirramycin A_1,[7] pikromycin,[8] kromycin,[8] and, to a lesser extent, the spiramycins,[9,10]



(1) $R^1 = R^2 = R^3 = R^4 = H$
(2) R^1 = Ac, $R^2 = R^3 = R^4 = H$
(3) $R^1 = R^2$ = Ac, $R^3 = R^4 = H$
(4) R^1 = EtCO, R^2 = Ac, $R^3 = R^4 = H$
(5) $R^1 = R^2 = H$, $R^3 = R^4$ = Ac
(6) $R^1 = R^4$ = Ac, $R^2 = R^3 = H$
(7) $R^1 = R^3 = R^4$ = Ac, $R^2 = H$
(8) $R^1 = R^2 = R^4$ = Ac, $R^3 = H$
(9) $R^1 = R^2 = R^3 = R^4$ = Ac
(10) $R^1 = R^2 = R^4 = H$, R^3 = EtCO
(11) R^1 = EtCO, $R^2 = R^3 = R^4 = H$
(12) $R^1 = R^2 = H$, $R^3 = R^4$ = EtCO
(13) $R^1 = R^2$ = EtCO, $R^3 = R^4 = H$
(14) $R^1 = R^3 = R^4$ = EtCO, $R^2 = H$
(15) $R^1 = R^2 = R^3 = R^4$ = EtCO
(16) $R^1 = R^2$ = Ac, R^3 = EtCO, $R^4 = H$

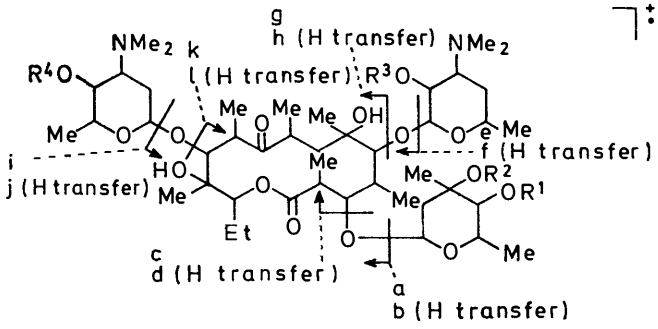


(17)

the magnamycins,[9,10] and the leucomycins.[11] We describe here our observations on the fragmentation patterns of the megalomicins,[1,12-14] a new group of macrolide antibiotics elaborated by *Micromonospora megalomicea* sp. n.

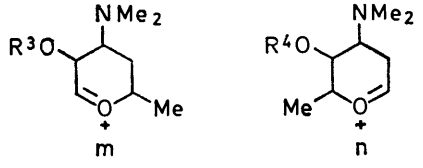
Extensive use of mass spectrometry was made during the elucidation of the structures of megalomicins A (1), B (2), C_1 (3), and C_2 (4). The principal fragmentations of these antibiotics, together with a number of selected acyl derivatives, are given in Table 1. In the case of megalomicin A (1), the compositions of the molecular ion and many of the major fragment ions were checked by high resolution measurements (Table 2).

In general the megalomicins exhibited the expected cleavages at the glycosidic linkages of the three sugar residues (Scheme 1). The glycosidic cleavages e—l of the desosamine and rhodosamine units were more



SCHEME 1

pronounced than the corresponding cleavages a—d of the mycarose residue, and the base peak of the spectrum



in general was due to the ions m and/or n. The presence of the ions m and n, and their high relative intensity

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TABLE 1

Megalomicin derivatives

Compound	Formula	M^+	a	b	c	d	e	f	g	h	i	j	k
(1)	$C_{44}H_{80}N_2O_{15}$	876(0·46)	731(0·12)	732(0·2)	715(0·1)	716(0·06)	718(0·3)	719(0·2)	702(0·28)	703(0·37)	718(0·3)	719(0·2)	702(0·28)
(2)	$C_{46}H_{82}N_2O_{16}$	918(0·25)	731(0·13)	732(0·21)	715(0·08)	716(0·06)	760(0·17)	761(0·15)	744(0·18)	745(0·17)	760(0·17)	761(0·15)	744(0·18)
(3)	$C_{48}H_{84}N_2O_{17}$	960(0·11)	731(0·10)	732(0·05)	715(0·14)	716(0·07)	802(0·15)	803(0·10)	786(0·23)	787(0·24)	802(0·15)	803(0·10)	786(0·23)
(4)	$C_{49}H_{86}N_2O_{17}$	974(0·1)	731(0·2)	732(0·2)	715(0·3)	716(0·06)	816(0·1)	817(0·07)	800(0·14)	801(0·14)	816(0·1)	817(0·07)	800(0·14)
(5) [16]	$C_{48}H_{84}N_2O_{17}$	960(0·56)	815(0·08)	816(0·11)	799(0·1)	800(0·25)	760(0·37)	761(0·14)	744(0·65)	745(0·5)	760(0·37)	761(0·14)	744(0·65)
(6)	$C_{48}H_{84}N_2O_{17}$	960(0·03)	773(0·04)	774(0·1)			802(0·03)	803(0·03)	786(0·04)	787(0·02)	760(0·04)	761(0·02)	744(0·1)
(7)	$C_{50}H_{86}N_2O_{18}$	1002(0·3)	815(0·1)	816(0·1)	799(0·1)	800(0·07)	802(0·2)	803(0·1)	786(0·4)	787(0·3)	802(0·2)	803(0·1)	786(0·4)
(8)	$C_{50}H_{86}N_2O_{18}$	1002(0·05)	773(0·05)	774(0·05)	757(0·06)	758(0·04)	844(0·05)	845(0·03)	828(0·06)	829(0·04)	802(0·07)	803(0·04)	786(0·12)
(9)	$C_{53}H_{88}N_2O_{19}$	1044(0·16)	815(0·05)		799(0·07)	800(0·05)	844(0·08)	845(0·05)	828(0·28)	829(0·21)	844(0·08)	845(0·05)	828(0·28)
(10)	$C_{47}H_{84}N_2O_{18}$	932(0·65)	787(0·1)	788(0·2)	771(0·1)		718(0·5)	719(0·2)	702(0·6)	703(0·3)	774(0·4)	775(0·3)	758(1·2)
(11)	$C_{47}H_{84}N_2O_{16}$	932(0·2)	731(0·25)	732(0·2)	715(0·2)	716(0·15)	774(0·4)	775(0·2)	758(0·65)	759(0·75)	774(0·4)	775(0·2)	758(0·65)
(12)	$C_{50}H_{88}N_2O_{17}$	988(0·08)	843(0·05)	844(0·09)	827(0·03)	828(0·02)	774(0·11)	775(0·05)	758(0·34)	759(0·27)	774(0·11)	775(0·05)	758(0·34)
(13)	$C_{50}H_{88}N_2O_{17}$	988(0·14)	731(0·09)	732(0·05)	715(0·13)	716(0·06)	830(0·19)	831(0·15)	814(0·22)	815(0·24)	830(0·19)	831(0·15)	814(0·22)
(14)	$C_{53}H_{92}N_2O_{18}$	1044(0·04)	843(0·03)	844(0·03)	827(0·03)	828(0·02)	830(0·09)	831(0·04)	814(0·27)	815(0·18)	830(0·09)	831(0·04)	814(0·27)
(15)	$C_{56}H_{98}N_2O_{19}$	1100(0·02)	843(0·03)	844(0·01)	827(0·04)	828(0·1)	886(0·07)	887(0·04)	870(0·23)	871(0·15)	886(0·07)	887(0·04)	870(0·23)
(16)	$C_{51}H_{88}N_2O_{18}$	1016(0·43)	787(0·15)	788(0·09)	771(0·11)	772(0·08)	802(0·14)	803(0·07)	786(0·2)	787(0·15)	858(0·14)	859(0·11)	842(0·46)
(17)	$C_{42}H_{73}N_2O_{15}$	831(0·33)		687(0·45)		671(0·12)	718(0·06)	719(0·03)	702(0·03)	703(0·02)			657(0·02)

Compound	Formula		b and e	b and f	b and g	b and h	b and i	b and j	b and k	b and l	m	n	o
(1)	$C_{44}H_{80}N_2O_{15}$	703(0·37)	574(0·3)	575(0·15)	558(0·9)	559(0·4)	574(0·3)	575(0·15)	558(0·9)	559(0·4)	158(100)	158(100)	801(0·75)
(2)	$C_{46}H_{82}N_2O_{16}$	745(0·17)	575(0·13)	575(0·05)	558(0·43)	559(0·22)	574(0·13)	575(0·05)	558(0·43)	559(0·22)	158(100)	158(100)	843(0·35)
(3)	$C_{48}H_{84}N_2O_{17}$	787(0·24)			558(0·29)	559(0·12)			558(0·29)	559(0·12)	158(100)	158(100)	885(0·18)
(4)	$C_{49}H_{86}N_2O_{17}$	801(0·14)	574(0·04)	575(0·02)	558(0·7)	559(0·1)	574(0·04)	575(0·02)	558(0·7)	559(0·1)	158(100)	158(100)	899(0·17)
(5) [16]	$C_{48}H_{84}N_2O_{17}$	745(0·05)	616(0·2)	617(0·8)	600(0·7)	601(0·38)	616(0·2)	617(0·08)	600(0·7)	601(0·38)	200(100)	200(100)	843(1·7)
(6)	$C_{48}H_{84}N_2O_{17}$	745(0·07)	616(0·08)	617(0·02)	600(0·07)	601(0·05)	574(0·05)	575(0·04)	558(0·4)	559(0·25)	158(53)	200(69)	843(0·2)
(7)	$C_{50}H_{86}N_2O_{18}$	787(0·3)	616(0·1)	617(0·03)	600(0·3)	601(0·18)	616(0·1)	617(0·03)	600(0·3)	601(0·18)	200(100)	200(100)	885(1·1)
(8)	$C_{50}H_{86}N_2O_{18}$	787(0·12)	616(0·02)	617(0·01)	600(0·05)	601(0·02)			558(0·16)	559(0·08)	158(62)	200(100)	885(0·19)
(9)	$C_{53}H_{88}N_2O_{19}$	829(0·21)	616(0·03)	617(0·01)	600(0·15)	601(0·06)	616(0·03)	617(0·01)	600(0·15)	601(0·06)	200(100)	200(100)	927(0·4)
(10)	$C_{47}H_{84}N_2O_{16}$	759(0·7)	574(0·6)	575(0·3)	558(0·5)	559(0·2)	630(0·5)	631(0·6)	614(1·4)	615(0·8)	214(60)	158(100)	857(1·7)
(11)	$C_{47}H_{84}N_2O_{16}$	759(0·75)	574(0·3)	575(0·2)	558(1·5)	559(0·7)	574(0·3)	575(0·2)	558(1·5)	559(0·7)	158(100)	158(100)	857(1·0)
(12)	$C_{50}H_{88}N_2O_{17}$	759(0·27)	630(0·6)	631(0·12)	614(0·82)	615(0·52)	630(0·33)	631(0·12)	614(0·82)	615(0·52)	214(60)	214(60)	857(0·51)
(13)	$C_{50}H_{88}N_2O_{17}$	815(0·24)	574(0·1)	575(0·04)	558(0·35)	559(0·13)	574(0·1)	575(0·04)	558(0·35)	559(0·13)	158(100)	158(100)	913(0·3)
(14)	$C_{53}H_{92}N_2O_{18}$	815(0·18)	630(0·08)	631(0·03)	614(0·23)	615(0·11)	630(0·08)	631(0·03)	614(0·23)	615(0·11)	214(100)	214(100)	913(0·44)
(15)	$C_{56}H_{98}N_2O_{19}$	871(0·15)	630(0·02)	631(0·01)	614(0·09)	615(0·03)	630(0·02)	631(0·01)	614(0·09)	615(0·03)	214(100)	214(100)	969(0·23)
(16)	$C_{51}H_{88}N_2O_{18}$	843(0·32)	574(0·02)	575(0·01)	558(0·13)	559(0·04)	630(0·06)	631(0·03)	614(0·31)	615(0·12)	214(55)	158(100)	941(0·55)
(17)	$C_{42}H_{73}N_2O_{15}$	658(0·08)	574(0·3)	575(0·1)	558(0·16)	559(0·06)						158(100)	756(0·4)

Compound	Formula	p	q	r	s and/or t	u	v	w	x	y	z	aa	bb
(1)	$C_{44}H_{80}N_2O_{15}$	657(0·55)	174(8)	174(8)	140(3)	87(14)	86(5)	71(30)	70(3·5)	98(18)	116(15)	114(8)	100(26)
(2)	$C_{46}H_{82}N_2O_{16}$	657(0·28)	174(7)	174(7)	140(4)	87(9)	86(4)	71(26)	70(3)	98(17)	116(13)	114(8)	100(27)
(3)	$C_{48}H_{84}N_2O_{17}$	657(0·28)	174(7)	174(7)	140(4)	87(7)	86(3)	71(20)	70(2)	98(16)	116(13)	114(7)	100(22)
(4)	$C_{49}H_{86}N_2O_{17}$	657(0·8)	174(8)	174(8)	140(4)	87(6)	86(3)	71(17)	70(2)	98(15)	116(16)	114(7)	100(20)
(5) [16]	$C_{48}H_{84}N_2O_{17}$	699(1·0)	216(6)	216(6)	140(9)	129(2) 87(6)	86(2)	71(21)	70(2)	98(16)	116(8)	156(9) 114(8)	100(23)
(6)	$C_{48}H_{84}N_2O_{17}$	657(0·3)	174(9)	216(2·5)	140(9)	87(13)	86(4)	71(36)	70(4)	98(26)	116(13)	156(12) 114(10)	100(33)
(7)	$C_{50}H_{86}N_2O_{18}$	699(0·7)	216(6)	216(6)	140(10)	129(3) 87(6)	86(4)	71(32)	70(4)	98(27)	116(9)	156(12) 114(10)	100(31)
(8)	$C_{50}H_{86}N_2O_{18}$	657(0·2)	174(19)	216(4)	140(8)	129(2) 87(8)	86(4)	71(24)	70(4)	98(35)	116(19)	156(15) 114(12)	100(48)
(9)	$C_{52}H_{88}N_2O_{19}$	699(0·75)	216(5)	216(5)	140(7)	129(2) 87(2)	86(1)	71(11)	70(1)	98(12)	116(5)	156(7) 114(6)	100(17)
(10)	$C_{47}H_{84}N_2O_{16}$	713(0·1)	230(3)	174(13)	140(25)	87(13)	86(7)	71(51)	70(4)	98(47)	116(19)	114(16)	100(45)
(11)	$C_{47}H_{84}N_2O_{16}$	657(0·9)	174(13)	174(13)	140(7)	87(9)	86(5)	71(27)	70(3)	98(25)	116(16)	114(11)	100(25)
(12)	$C_{50}H_{88}N_2O_{17}$	713(0·8)	230(3·7)	230(3·7)	140(8·3)	87(8)	86(2)	71(13)	70(2)	98(16)	116(7)	170(4·7) 114(6)	100(16)
(13)	$C_{50}H_{88}N_2O_{17}$	657(0·3)	174(8)	174(8)	140(4)	87(7)	86(4)	71(24)	70(3)	98(21)	116(16)	114(8)	100(28)
(14)	$C_{53}H_{92}N_2O_{18}$	713(0·4)	230(5)	230(5)	140(10)	87(6)	86(4)	71(35)	70(5)	98(23)	116(8)	170(10) 114(11)	100(26)
(15)	$C_{56}H_{96}N_2O_{19}$	713(0·34)	230(6)	230(6)	140(12)	87(3)	86(2)	71(18)	70(2)	98(19)	116(8)	170(8) 114(9)	100(26)
(16)	$C_{51}H_{88}N_2O_{16}$	713(0·46)	230(2)	174(5)	140(16)	87(4)	86(3)	71(21)	70(2)	98(22)	116(10)	114(9)	100(26)
(17)	$C_{42}H_{73}N_2O_{15}$	612(0·8)		174(4)		87(17)	86(8)	71(64)	70(5)			114(7)	100(47)

Compound	Formula	cc	dd	ee	ff	gg	hh	ii	jj	kk and/or ll	Miscellaneous
(1)	$C_{44}H_{80}N_2O_{15}$	145(4)	127(5)	109(2·5)	748(0·77)	604(0·35)	444(0·6)	426(0·3)	818(0·04)	791(0·2)	
(2)	$C_{46}H_{82}N_2O_{16}$	187(4)	169(4)	109(12)	790(0·32)	604(0·13)	444(0·24)	426(0·37)	860(0·07)	833(0·14)	
(3)	$C_{48}H_{84}N_2O_{17}$	229(3)	169(15)	109(27)	832(0·31)		444(0·09)	426(0·3)	902(0·01)	875(0·07)	$M-60$: 900(0·02)
(4)	$C_{49}H_{86}N_2O_{17}$	243(3)	183(12)	109(27)	846(0·19)		444(0·06)	426(0·4)	916(0·02)	889(0·05)	$M-60$: 914(0·02); $M-74$: 900(0·09)
(5) [16]	$C_{48}H_{84}N_2O_{17}$	145(2)	127(4)	109(3)			486(0·16)	468(0·06)	902(0·2)	875(0·05)	$M-60$: 900(0·12); $M-60-42$; 858(0·85)
(6)	$C_{48}H_{84}N_2O_{17}$	187(4)	169(3)	109(14)	832(0·16)	646(0·1)	486(0·25)	468(0·12)	902(0·011)	875(0·01)	$M-60$: 900(0·02); $M-60-42$; 858(0·04)
(7)	$C_{50}H_{86}N_2O_{18}$	187(4)	169(2·5)	109(12)			486(0·2)	468(0·1)	944(0·15)	917(0·05)	$M-60$: 942(0·08); $M-60-42$; 900(0·5)
(8)	$C_{50}H_{86}N_2O_{18}$	229(3)	169(26)	109(56)	874(0·15)	646(0·04)	486(0·07)	468(0·08)	986(0·01)	959(0·01)	$M-60$: 942(0·02); $M-60-42$; 900(0·01)
(9)	$C_{53}H_{88}N_2O_{19}$	229(2)	169(9)	109(16)			486(0·03)	468(0·06)	986(0·05)	959(0·02)	$M-60$: 984(0·01); $M-60-42$; 942(0·19)
(10)	$C_{47}H_{84}N_2O_{16}$	145(7)	127(10)	109(3)			444(2·6)	426(1·0)	874(0·15)	847(0·1)	$M-56$: 801(0·6)
(11)	$C_{47}H_{84}N_2O_{16}$	201(12)	183(7)	109(16)	804(1·1)	604(0·4)	444(0·6)	426(1·0)	874(0·08)	847(0·15)	$M-56$: 801(0·6)
(12)	$C_{50}H_{88}N_2O_{17}$	145(15)	127(4·5)	109(26)			500(0·22)	482(0·07)	930(0·02)	903(0·01)	
(13)	$C_{50}H_{88}N_2O_{17}$	257(3)	183(18)	109(48)	860(0·45)		444(0·1)	426(0·21)	930(0·03)	903(0·1)	
(14)	$C_{53}H_{92}N_2O_{18}$	201(3)	183(1·5)	109(9)			500(0·07)	482(0·1)	986(0·03)		
(15)	$C_{56}H_{96}N_2O_{19}$	257(3)	183(14)	109(33)			500(0·01)	482(0·04)	1042(0·01)		
(16)	$C_{51}H_{88}N_2O_{18}$	229(4)	169(19)	109(31)			444(0·08)	426(0·2)	958(0·1)	931(0·07)	$M-60$: 956(0·04)
(17)	$C_{42}H_{73}N_2O_{15}$	145(6)	127(9)	109(7)			444(0·3)	426(0·1)	773(0·04)	746(0·14)	mm: 748(0·19); mm − **18**: 730(0·17); mm − **b** 604(0·8); mm − k: 574(0·3); **nn**: 115(11)

compared with the remainder of the peaks affords a ready method of determining the compositions of any amino-sugar systems in a macrolide antibiotic. In the case of megalomicin A (1), the formation of one high intensity peak at *m/e* 158 suggested that both amino-sugar residues had the same composition, giving rise to the $C_8H_{16}NO_2$ ions, which constituted the base peak in

TABLE 2

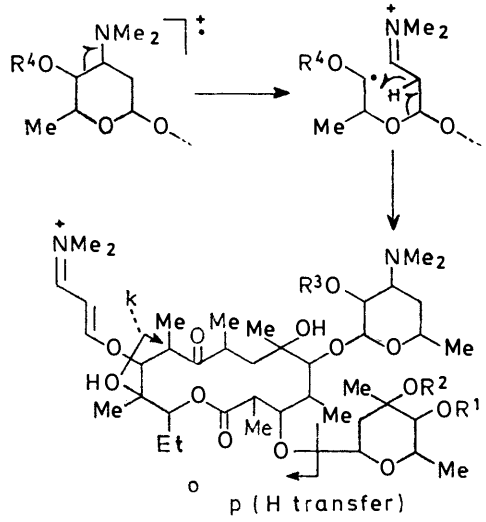
High resolution mass spectral data

Compound	Ion(s)	Calc.	Obs.
(1) [a]	M^+	876·5557	876·5496
	e and i	718·4375	718·4353
	f and j	719·4453	719·4409
	g and k	702·4426	702·4395
	h and l	703·4504	703·4468
	b and g; b and k	558·3640	558·3640
	b and h; b and l	559·3718	559·3748
	o	801·5110	801·5088
	ff	748·4482	748·4402
	kk and ll	791·4902	791·4868
	hh	444·2959	444·2913
	cc	145·0864	145·0886
	dd	127·0759	127·0765
	ee	109·0653	109·0666
	q and r	174·1130	174·1131
	m and n	158·1181	158·1166
	s and t	140·1075	140·1057
	y	98·0970	98·0979
	aa	114·0919	114·0928
	z	116·0711	116·0736
	bb	100·0762	100·0759
	u	87·0684	87·0688
	v	86·0606	86·0622
	w	71·0735	71·0732
	x	70·0657	70·0666
(37) [b]	M^+	539·346	539·345
	jj''''	481·304	481·301
	e''''	381·228	381·225
	g''''	365·232	365·229
	q	174·113	174·113
	m	158·118	158·118
(40) [b]	M^+	623·367	623·368
	e''''	423·238	423·235
	g''''	407·243	407·243
	ww	347·222	347·221
	m	200·129	200·131
	s	140·108	140·107

[a] Measured on an A.E.I. MS902B spectrometer. [b] Measured on a JEOL JMS-01SC spectrometer.

the spectrum. Acylation of the hydroxy-groups in each of the amino-sugars caused the base peak to shift to *m/e* 200 in the case of the acetates, and to *m/e* 214 in the case of the propionates. The glycosidic cleavages e—l in the high mass region also shifted by 42 and 56 mass units, respectively. Where only one of the hydroxy-groups in either of the amino-sugars was acylated, two intense peaks at *m/e* 158 and 200 (acetate) or 214 (propionate) were formed, the latter being of lower relative intensity owing to partial conversion into the ions *m/e* 158 by loss of keten or methylketen for the acetates and propionates, respectively. In those derivatives of the megalomicins where the acyl groups were located solely in the mycarose unit, the base peak in each case was at *m/e* 158, with the appropriate high mass cleavages e—l. The foregoing fragmentations afford a convenient means of locating various acyl groups in three specific areas of the molecule, namely the amino-sugar residues, the mycarose unit, and the aglycone. Peaks were also observed for successive cleavages involving mycarose and either desosamine or rhodosamine, and are given in Table 1.

One of the principal fragmentations in the rhodosamine unit is outlined in Scheme 2. In the case of megalomicin A (1) this loss of 75 mass units from the molecular ion to give the ion o indicated that rhodosamine was a 2,3,6-trideoxy-3-dimethylamino-sugar, a conclusion confirmed by isolation and chemical degradation of the D-rhodosamine.[12,14] The formation of the ion o made it



SCHEME 2

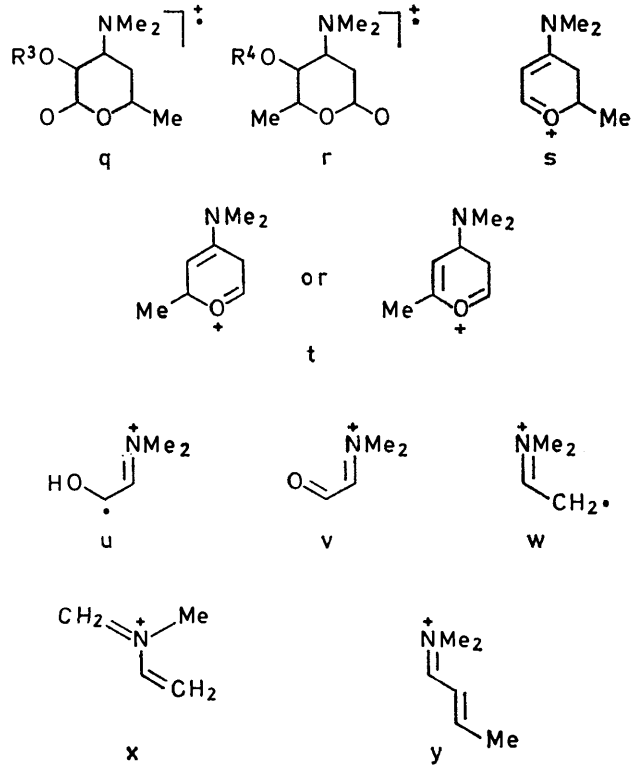
possible not only to locate an acyl group in the amino-sugars as already described, but also to determine which of the two amino-sugars contained the acyl substituent. This provided a convenient method for locating the acetyl group in the rhodosamine in the esters (6) and (8), and the propionyl group in the desosamine in the esters (10) and (16). In the case of the acetyl derivatives of the megalomicins it was possible to confirm the foregoing assignments, as well as all of those indicated in Table 1, from the chemical shifts of the acetyl groups in the n.m.r. spectra.[1,15,16] This was not possible with the propionates. The ion o was found to undergo further cleavage to give the ion k by loss of 99 mass units, and also the ion p by loss of the mycarose unit accompanied by a hydrogen atom transfer.

The low mass region of the spectra showed characteristic fragment ions from the expected cleavages of desosamine and rhodosamine.[14] In addition to the base peak due to m and n, peaks were also present due to fragments q and r. The further loss of water or alkanecarboxylic acid (acetic or propionic) from m and n gave rise to a peak at *m/e* 140 due to the fragments s

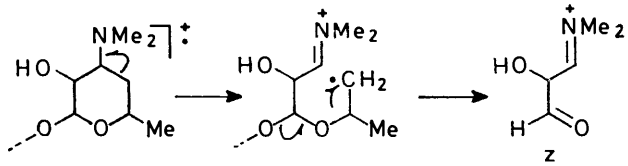
[15] R. S. Jaret, A. K. Mallams, H. Reimann, and H. F. Vernay, unpublished observations.

[16] Compounds (11), (13), and (15) were prepared under the direction of H. Reimann; manuscript in preparation.

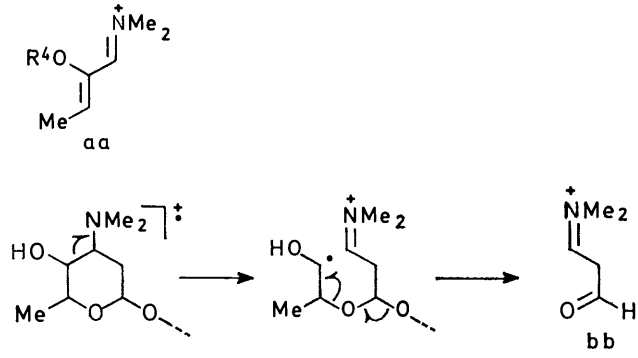
and t. The expected cleavages of the amino-sugars led to the formation of ions u—x in which the charge was



stabilised on the nitrogen atom.[14] In addition, desosamine gave rise to fragment ions y [14] and z, and rhodosamine gave ions aa [14] and bb, respectively. The formation of the ions z and bb may be postulated to occur as



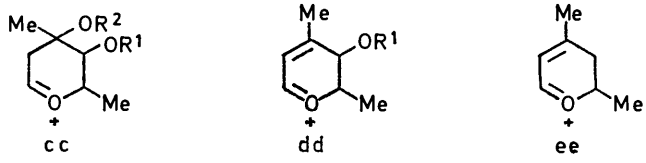
SCHEME 3



SCHEME 4

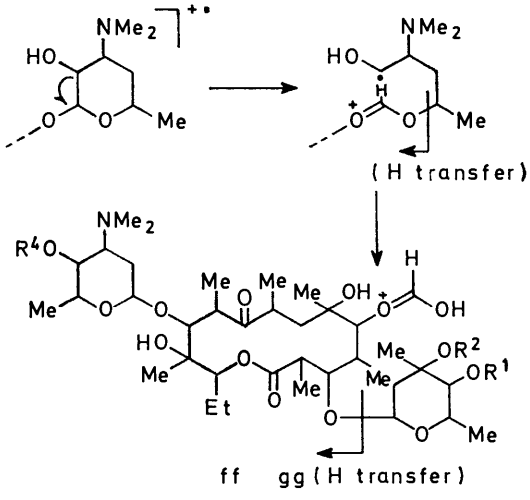
outlined in Schemes 3 and 4, respectively, with stabilisation of the charge on the nitrogen atom in both instances.

In those derivatives of the megalomicins where the acyl groups were located in the mycarose unit, this was evident from the high mass ions arising from the glycosidic cleavages a—d. In those examples where the mycarose contained one acyl substituent, or where the mycarose contained mixed acyl substituents, it was possible, by locating peaks due to the fragment ions cc, dd, and ee in the low mass region of the spectrum, to



assign the monoacyl or mixed acyl substituents to the 3- or 4-positions, respectively, in the mycarose unit. This proved to be a useful way of locating the acetyl group at C-4 in megalomicin B (2), and for locating the acetyl group at C-3′ and the propionyl group at C-4′ in megalomicin C_2 (4). Both these conclusions were checked by n.m.r. measurements and by isolation and degradation of the appropriate mycarose units from structures (2) and (4).[1]

In those megalomicin derivatives where the desosamine was not acylated, a fragment ion was observed corresponding to $M^+ - 128$, which could arise from the ion ff (Scheme 5). Peaks corresponding to further



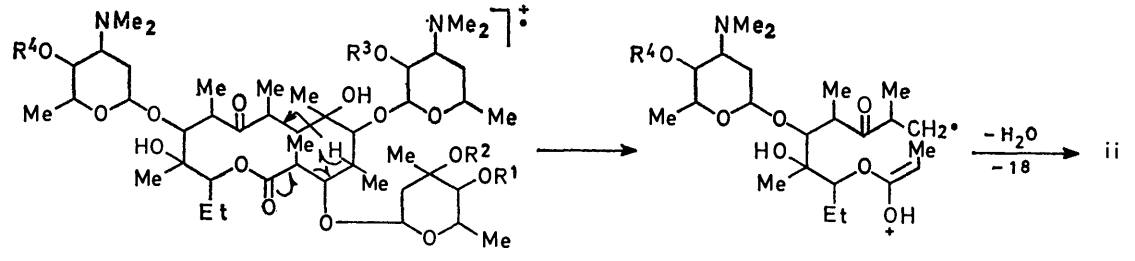
SCHEME 5

glycosidic cleavage of the mycarosyl residue leading to the formation of the ions gg from the ions ff were also observed (Scheme 5).

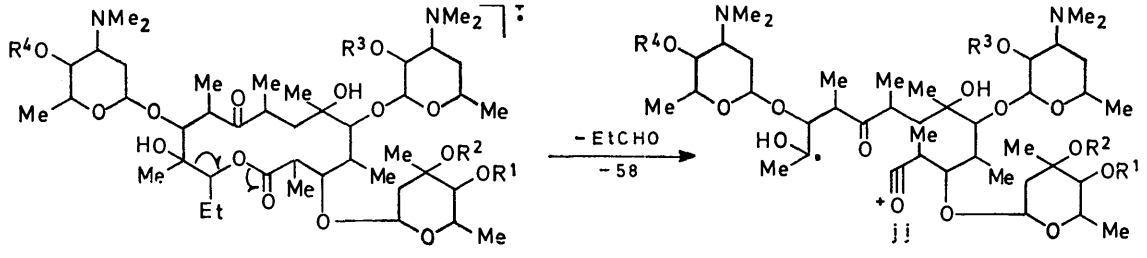
The megalomicins underwent several interesting cleavages in the aglycone. One of these gave rise to what is postulated to be the ion hh, which could arise as a result of a McLafferty rearrangement at the lactone carbonyl group accompanied by a cleavage of the C(6)–C(7) bond adjacent to the tertiary C-6 (Scheme 6). In those megalomicin derivatives where the rhodosamine

was acylated the fragment hh shifted to a higher *m/e* value by 42 mass units in the case of an acetate and by 56 mass units in the case of a propionate. The presence of the ion hh provided additional evidence for locating the rhodosamine unit between C-7 and C-13 in the aglycone, and also for determining the location of a monoacyl substituent in either the desosamine or cleavage adjacent to the tertiary C-12 would constitute a favourable cracking pattern leading to the formation of ion jj.

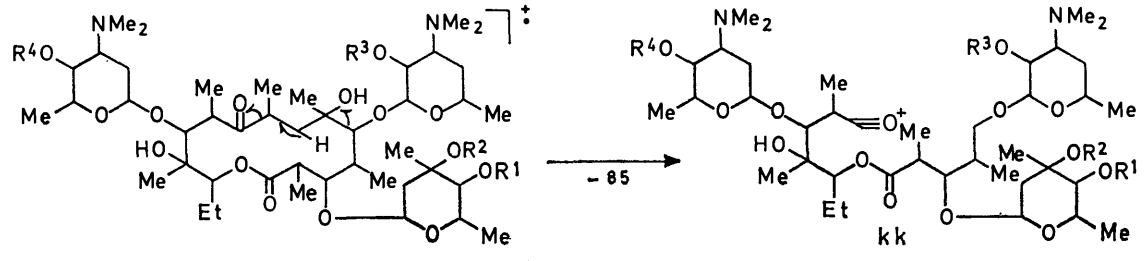
A medium-intensity peak in the high mass region of the spectra of the megalomicins corresponding to a loss of a C_5H_9O unit ($M^+ - 85$) could arise either as a result of cleavage α to the 9-oxo-group and adjacent to the



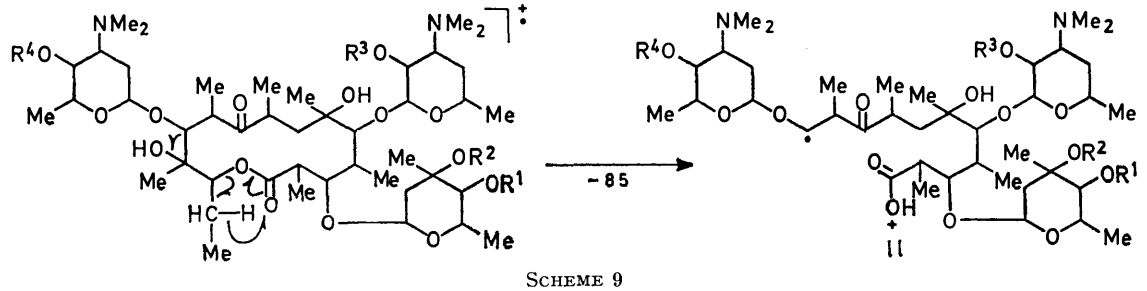
SCHEME 6



SCHEME 7



SCHEME 8



SCHEME 9

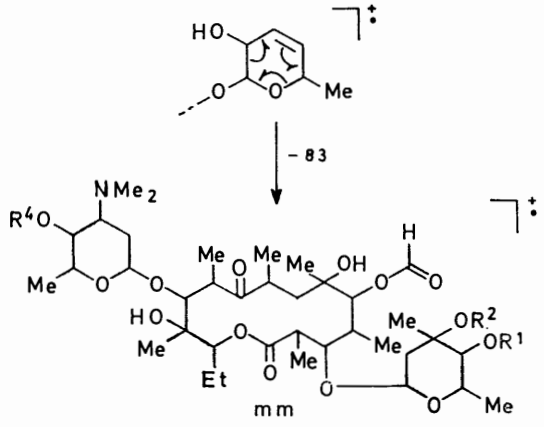
rhodosamine units. The fragment ion hh also lost a molecule of water to give the ion ii.

The megalomicins also gave a peak in the high mass region corresponding to the loss of propionaldehyde ($M^+ - 58$), leading to the ion jj (Scheme 7). Cleavage next to the lactone carbonyl group with stabilisation of the charge on the carbonyl oxygen atom, together with tertiary C-6 with hydrogen atom transfer, leading to ion kk (Scheme 8), or *via* a McLafferty rearrangement at the lactone system, accompanied by cleavage adjacent to the tertiary C-12 to give ion ll (Scheme 9). Both processes appear to be favourable on mass spectral grounds.

In general, the acetyl derivatives of the megalomicins

lost acetic acid and/or keten from the molecular ion (Table 1), whereas the propionates lost propionic acid, and/or methylketen.

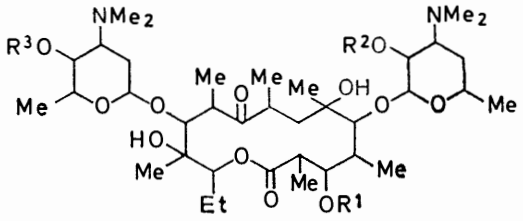
The mass spectrum of 3''-de(dimethylamino)-3'',4''-didehydromegalomicin A (17), in which the desosamine residue had been deaminated selectively,[17] confirmed many of the foregoing assignments. The glycosidic cleavages associated with the mycarosyl unit by paths b † and d † accompanied by ions cc, dd, and ee in the low mass region were observed as in the case of the other megalomicin derivatives. Cleavage of the deaminated desosamine system gave rise to the ions e—h † by the processes already outlined. The base peak in the spectrum was again found to be due to the ion n at *m/e* 158, derived in this instance solely from the rhodosamine unit. The deaminated desosamine gave rise to a high mass ion mm by a retro-Diels-Alder fission (Scheme 10)



SCHEME 10

as well as a low mass ion nn at *m/e* 113. The ion mm underwent further losses of a water molecule (mm — 18), a mycarosyl unit (mm — b), and a rhodosaminyl unit (mm — k) giving rise to ions at *m/e* 730, 604, and 574, respectively. The fragment ions aa and z, associated with the rhodosamine unit, were present in the mass spectrum of (17), while the ions y and bb, derived from amine-induced cleavages in the desosamine residue, were absent in the spectrum. The expected ions u—x [14] common to both amino-sugars were observed in the spectrum of (17). As in the case of the megalomicins where $R^4 = H$, the fragments at *m/e* 444 (hh) and 426 (ii) were again observed in the mass spectrum of (17), further supporting the fact that these ions contained the rhodosamine unit. Losses of 58 mass units to give the fragment jj † and of 85 mass units to give ions kk † and ll † were also observed in the spectrum of (17).

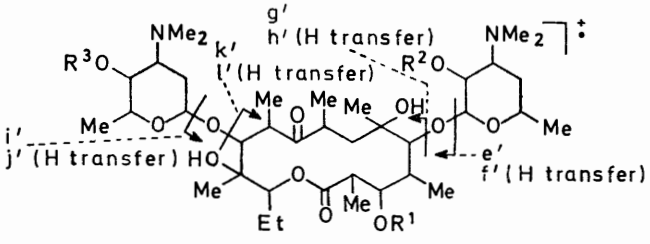
In the course of the degradation of the megalomicins, mild aqueous acidic hydrolysis was found to cause selective hydrolysis of the mycarose giving megalalosamine (18).[1,13] A number of derivatives (19)—(29) of megalalosamine (18) were prepared,[17] and some of the more important fragment ions from these compounds are given in Table 3. In general the megalalosamines exhibited similar fragmentations to those encountered



(18) $R^1 = R^2 = R^3 = H$
(19) $R^1 = Ac$, $R^2 = R^3 = H$
(20) $R^1 = R^2 = H$, $R^3 = Ac$
(21) $R^1 = H$, $R^2 = R^3 = Ac$
(22) $R^1 = R^2 = R^3 = Ac$
(23) $R^1 = EtCO$, $R^2 = R^3 = Ac$
(24) $R^1 = Bu^iCO$, $R^2 = R^3 = Ac$
(25) $R^1 = EtCO$, $R^2 = R^3 = H$
(26) $R^1 = R^3 = EtCO$, $R^2 = H$
(27) $R^1 = R^2 = R^3 = EtCO$
(28) $R^1 = H$, $R^2 = R^3 = Bz$
(29) R^1 = tetrahydropyranyl, $R^2 = R^3 = Bz$

with the megalomicins, with the exception of the mycarose-derived ions which were absent in the former.

Fragmentation at the glycosidic bonds in the megalalosamines gave rise to the ions e'—l',‡ and the base peak in the spectrum was due to ions m and/or n, in general. Acylation of the hydroxy-groups in the amino-sugars again caused the appropriate shifts in the *m/e* values of the ions e'—l', and also caused the base peak mass number to increase by the expected values for the various acyl substituents. In the case of the triacyl derivatives it was possible by studying the *m/e* values of the foregoing ions to locate the various acyl substituents in the amino-sugar residues or in the aglycone, respectively. Further distinctions between the location of a particular acyl substituent in the desosamine or rhodosamine units could then be made from the *m/e* values of the ion o'. A 10 eV scan of megalalosamine revealed the presence of metastable peaks at *m** 589·7 corresponding to the transition $M^+ \longrightarrow$ o' and also *m** 473·9 corresponding



SCHEME 11

to o' $\longrightarrow$ k'. These metastable peaks afforded evidence for the fact that the ion k' was produced not only by a single-stage glycosidic cleavage (Scheme 11), but also

† The analogous ion in which the desosamine unit had been deaminated.

‡ Where analogous fragmentations have been discussed for the megalomicins these are indicated by the same alphabetical symbol followed by a prime notation in the case of the megalalosamines, a double prime notation in the case of the dihydro-derivatives, a triple prime notation, in the case of the erythromycins, and a quadruple prime notation for the erythralosamines.

[17] A. K. Mallams and H. F. Vernay, unpublished observations.

by a two-stage cleavage involving the ion o′. The presence of the fragments hh and ii in the megalalosamines also proved useful in locating an acyl substituent if it was present in the rhodosamine unit, although these not acylated. The loss of propionaldehyde from the molecular ion to give ion jj′ was observed in most of the derivatives, and ions kk′ and ll′ were also present in the mass spectra of the megalalosamines. Losses of acetic,

TABLE 3

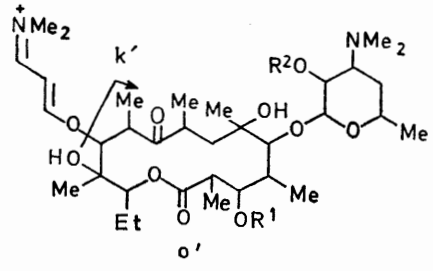
Megalalosamine derivatives

Compd	Formula	M^+	e′	f′	g′	h′	i′	j′	k′	l′	m	n
(18)	$C_{37}H_{68}N_2O_{12}$	732(0·48)	574(0·57)	575(0·33)	558(1·26)	559(1·05)	574(0·57)	575(0·33)	558(1·26)	559(1·05)	158(100)	158(100)
(19)	$C_{39}H_{70}N_2O_{13}$	774(1·2)	616(1·0)	617(0·6)	600(1·3)	601(2·0)	616(1·0)	617(0·6)	600(1·3)	601(2·0)	158(100)	158(100)
(20)	$C_{39}H_{70}N_2O_{13}$	774(0·65)	616(1·3)	617(0·5)	600(0·3)	601(0·35)	574(0·5)	575(0·2)	558(4·3)	559(3·3)	158(99)	200(100)
(21)	$C_{41}H_{72}N_2O_{14}$	816(0·17)	616(0·6)	617(0·2)	600(1·0)	601(0·8)	616(0·6)	617(0·2)	600(1·0)	601(0·8)	200(100)	200(100)
(22)	$C_{43}H_{74}N_2O_{15}$	858(0·74)	658(0·84)	659(0·38)	642(1·0)	643(1·8)	658(0·84)	659(0·38)	642(1·0)	643(1·8)	200(100)	200(100)
(23)	$C_{44}H_{76}N_2O_{15}$	872(0·65)	672(0·22)	673(0·88)	656(1·17)	657(2·08)	672(0·22)	673(0·88)	656(1·17)	657(2·08)	200(100)	200(100)
(24)	$C_{46}H_{80}N_2O_{15}$	900(0·4)	700(0·55)	701(0·25)	684(0·75)	685(1·15)	700(0·55)	701(0·25)	684(0·75)	685(1·15)	200(100)	200(100)
(25)	$C_{40}H_{72}N_2O_{13}$	788(0·18)	630(0·25)	631(0·12)	614(0·28)	615(0·44)	630(0·25)	631(0·12)	614(0·28)	615(0·44)	158(100)	158(100)
(26)	$C_{43}H_{76}N_2O_{14}$	844(1·5)	686(0·53)	687(0·4)	670(0·3)	671(0·4)	630(0·95)	631(0·6)	614(2)	615(4)	158(100)	214(67)
(27)	$C_{46}H_{80}N_2O_{15}$	900(1·2)	686(1)	687(0·5)	670(1·5)	671(2·6)	686(1)	687(0·5)	670(1·5)	671(2·6)	214(100)	214(100)
(28)	$C_{51}H_{76}N_2O_{14}$	940(4·0)	678(0·9)	679(0·4)	662(7·0)	663(8·8)	678(0·9)	679(0·4)	662(7·0)	663(8·8)	262(94)	262(94)
(29)	$C_{56}H_{84}N_2O_{15}$	1024(0·015)	762(0·13)	763(0·05)	746(0·07)	747(0·05)	762(0·13)	763(0·05)	746(0·07)	747(0·05)	262(33)	262(33)
(30)	$C_{35}H_{61}NO_{12}$	687(0·55)	574(0·59)	575(0·22)			529(0·02)	530(0·02)				158(100)

Compd	Formula	o′	q	r	s and/or t	u	v	w	x	y	z	aa
(18)	$C_{37}H_{68}N_2O_{12}$	657(1·12)	174(7·5)	174(7·5)	140(2·5)	87(12)	86(8)	71(45)	70(6·5)	98(20)	116(16)	114(8)
(19)	$C_{39}H_{70}N_2O_{13}$	699(2·5)	174(9·3)	174(9·3)	140(6)	87(10)	86(7)	71(82)	70(26)	98(28)	116(15)	114(10)
(20)	$C_{39}H_{70}N_2O_{13}$	657(3·9)	174(25)	216(6)	140(10)	129(4)	86(10)	71(12)	70(8)	98(33)	116(30)	156(26)
						87(19)						114(17)
(21)	$C_{41}H_{72}N_2O_{14}$	699(0·9)	216(4)	216(4)	140(10)	129(2·5)	86(4)	71(36)	70(6)	98(28)	116(13)	156(11)
						87(5)						114(9)
(22)	$C_{43}H_{74}N_2O_{15}$	741(4·4)	216(6)	216(6)	140(8)	129(2)	86(4)	71(35)	70(5)	98(21)	116(9)	156(11)
						87(5)						114(10)
(23)	$C_{44}H_{76}N_2O_{15}$	755(6·7)	216(6)	216(6)	140(9)	129(3)	86(3)	71(29)	70(4)	98(20)	116(10)	156(14)
						87(3·8)						114(11)
(24)	$C_{46}H_{80}N_2O_{15}$	783(2·1)	216(6·5)	216(6·5)	140(10)	129(2·7)	86(4)	71(31)	70(3·5)	98(20)	116(10)	156(16)
						87(5)						114(12)
(25)	$C_{40}H_{72}N_2O_{13}$	713(0·57)	174(6)	174(6)	140(3)	87(10)	86(6)	71(11)	70(4)	98(19)	116(15)	114(8)
(26)	$C_{43}H_{76}N_2O_{14}$	713(4)	174(7)	230(4)	140(15)	87(17)	86(13)	71(76)	70(18)	98(52)	116(30)	170(22)
												114(31)
(27)	$C_{46}H_{80}N_2O_{15}$	769(4)	230(5)	230(5)	140(21)	87(7)	86(7)	71(46)	70(11)	98(46)	116(15)	170(14)
												114(22)
(28)	$C_{51}H_{76}N_2O_{14}$	761(7·1)	278(6)	278(6)	140(30)	191(3)		71(90)	70(12)	98(56)	116(15)	218(27)
												114(14)
(29)	$C_{56}H_{84}N_2O_{15}$	845(0·08)	278(2·7)	278(2·7)	140(15)	87(5)	86(9)	71(39)	70(15)	98(22)	116(6)	218(11)
												114(8)
(30)	$C_{35}H_{61}NO_{12}$	612(1·52)		174(5)	140(2)	87(6)	86(10)	71(77)	70(7)			114(9)

Compd	Formula	bb	ff′	hh	ii	jj′	kk′ and/or ll′	Miscellaneous
(18)	$C_{37}H_{68}N_2O_{12}$	100(30)	604(1·06)	444(0·25)	426(0·21)	674(0·04)	647(0·33)	
(19)	$C_{39}H_{70}N_2O_{13}$	100(27)	646(1·1)	444(0·2)	426(0·2)	716(0·15)	689(0·5)	$M-60$: 714(0·2)
(20)	$C_{39}H_{70}N_2O_{13}$	100(62)	646(2·1)	486(0·2)		716(0·11)	689(0·55)	$M-60$: 714(0·34)
(21)	$C_{41}H_{72}N_2O_{14}$	100(28)		486(0·1)	468(0·1)		731(0·04)	$M-60$: 756(0·05); $M-60-42$: 714(0·25)
(22)	$C_{43}H_{74}N_2O_{15}$	100(30)		486(0·06)	468(0·08)	800(0·26)	773(0·15)	$M-60$: 798(0·41)
(23)	$C_{44}H_{76}N_2O_{15}$	100(31)		486(0·1)		814(0·27)	787(0·15)	$M-60$: 812(0·4); $M-60-42$: 770(1·0); $M-74$: 798(0·37)
(24)	$C_{46}H_{80}N_2O_{15}$	100(35)		486(0·4)	468(0·09)	842(0·16)	815(0·1)	$M-60$: 840(0·2); $M-60-42$: 798(0·65); $M-102$: 798(0·65)
(25)	$C_{40}H_{72}N_2O_{13}$	100(32)	660(0·23)	444(0·04)	426(0·12)		703(0·1)	$M-74$: 714(0·25)
(26)	$C_{43}H_{76}N_2O_{14}$	100(71)	716(1·2)	500(0·3)	482(0·23)		759(0·43)	$M-74$: 770(1·2)
(27)	$C_{46}H_{80}N_2O_{15}$	100(47)					815(0·18)	$M-74$: 826(0·95)
(28)	$C_{51}H_{76}N_2O_{14}$	100(52)		548(0·35)			855(1·5)	$M-122$: 818(0·6)
(29)	$C_{56}H_{84}N_2O_{15}$	100(22)		548(0·1)	530(0·14)		939(0·07)	$M-122$: 902(0·01); a′: 939(0·07); c′: 923(0·01); oo: 85(19);
(30)	$C_{35}H_{61}NO_{12}$	100(50)		444(0·07)	426(0·1)	629(0·08)	602(0·37)	mm′: 603(0·27); nn: 113(8)

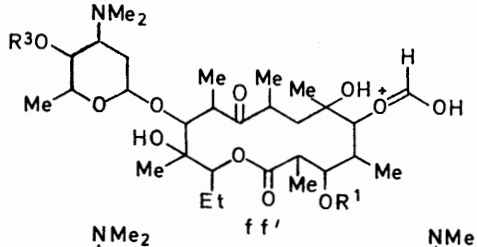
ions were of low intensity compared with o. In the case of the acetyl derivatives of the megalalosamines the location of the various acetyl groups was checked by observing the chemical shifts of these groups in the n.m.r. spectra,[17] the assignments were in complete



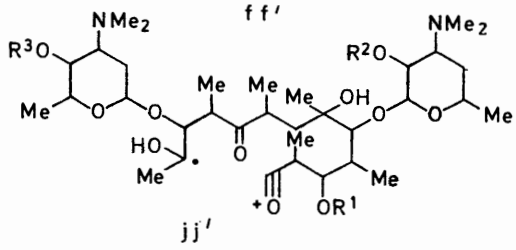
o′

agreement. The low mass region of the spectra showed the typical fragment ions q—z, aa, and bb derived from the amino-sugar systems. As in the case of the megalomicins the fragment ff′ was observed, due to the cleavage of the desosamine in derivatives where the latter was

propionic, and benzoic acids from the molecular ion, as well as combinations thereof, including the loss of keten



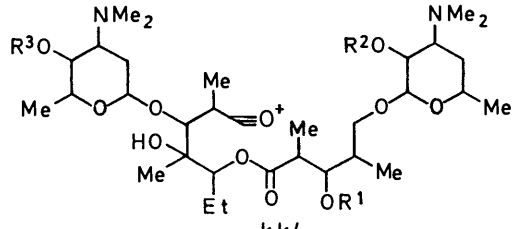
ff′



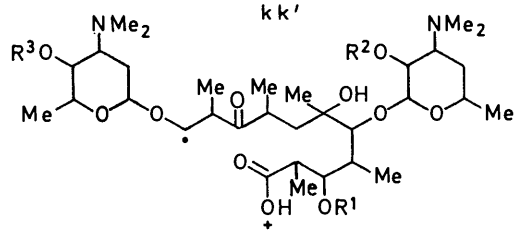
jj′

in a number of acetates, were apparent in the spectra of the acylmegalalosamines.

The mass spectrum of 2′,4″-di-*O*-benzoyl-3-tetrahydropyranyloxymegalalosamine (29) showed a close similarity to the spectra of the megalomicins, with glycosidic cleavages of the tetrahydropyranyl group giving rise to ions a′, c′, and oo. The spectrum of 3′-de(dimethylamino)-3′,4′-didehydromegalalosamine (30) showed the

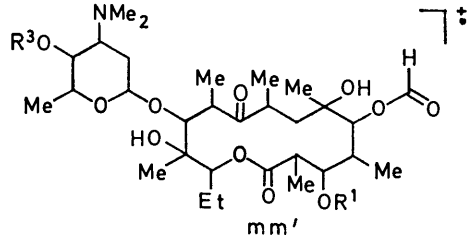


kk′

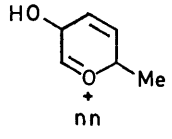


ll′

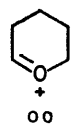
typical glycosidic cleavages related to the rhodosamine while lacking those of the desosamine. The deaminated desosamine unit underwent glycosidic cleavages to give the ions e′,† f′,† and nn, and also a retro-Diels–Alder



mm′



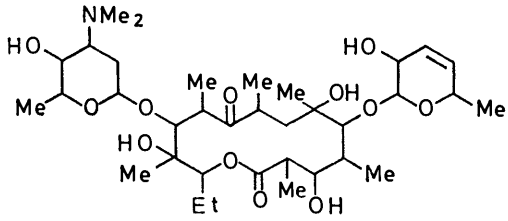
nn



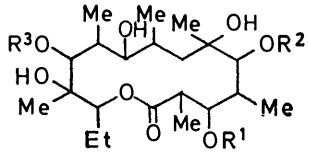
oo

fission to give the fragment ion mm′. The cleavages of the aglycone of (30) produced the ions hh, ii, and jj′—ll′ † as expected.

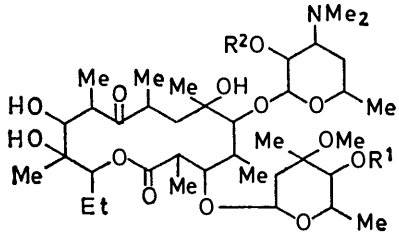
The major fragment ions in the mass spectra of 9,*O*(9)-dihydromegalomicin A (31),[1] 9,*O*(9)-dihydromegalalosamine (32),[1] and 5-β-D-desosaminyloxy-9,*O*(9)-dihydroerythronolide (33)[1,18] are listed in Table 4. Glycosidic cleavages of the sugar residues gave rise to the ions a″—l″ as in the case of the megalomicins and megalalosamines. The base peaks were again at *m/e* 158, due to the amino-sugar ions m and n. As in the 9-oxo-series, the loss of 75 mass units from the rhodosamine unit gave ion o″ in the spectra of (31) and (32), and this ion fragmented further to give k″ with an attendant loss of 99 mass units. The ion o″ was not observed in the spectrum of (33) as the latter contained



(30)

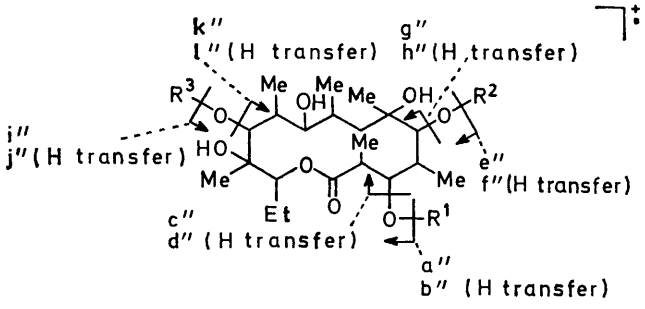


(31) R^1 = α-L-mycarosyl, R^2 = β-D-desosaminyl, R^3 = β-D-rhodosaminyl
(32) R^1 = H, R^2 = β-D-desosaminyl, R^3 = β-D-rhodosaminyl
(33) R^1 = R^3 = H, R^2 = β-D-desosaminyl



(34) R^1 = R^2 = H
(35) R^1 = H, R^2 = Ac
(36) R^1 = R^2 = Ac

no rhodosamine sugar. Fragmentation of the desosamine in (31) and (32) gave rise to the ion ff″ in each case. The low-mass regions of the spectra contained peaks due



SCHEME 12

to the ions q—bb, formed by fragmentation of the desosamine and rhodosamine units (where the latter was present) and also, in the case of (31), the ions cc—ee produced by fragmentation of the mycarose residue. The ions analogous to hh—ii formed in the megalomicins and megalalosamines were not observed in the

† The analogous ion in which the desosamine unit has been deaminated.

[18] M. V. Sigal, P. F. Wiley, K. Gerzon, E. H. Flynn, U. C. Quarck, and O. Weaver, *J. Amer. Chem. Soc.*, 1956, **78**, 388.

TABLE 4

9-Hydroxy-analogues

Compd	Formula	M^+	a''	b''	c''	d''	e''	f'	g''	h''	i''	j''
(31)	$C_{44}H_{82}N_2O_{15}$	878(0·02)	733(0·05)	734(0·1)	717(1)	718(0·05)	720(1)	721(1)	704(3)	705(2)	720(1)	721(1)
(32)	$C_{37}H_{70}N_2O_{12}$	734(0·4)					576(2·1)	577(1·4)	560(3·5)	561(1·4)	576(2·1)	577(1·4)
(33)	$C_{29}H_{55}NO_{10}$	577(2·6)							403(1·0)	404(0·4)		

Compd	Formula	k''	l''	m	n	o[a]	q	r	s and/or t	u	v	w
(31)	$C_{44}H_{82}N_2O_{15}$	704(3)	705(2)	158(100)	158(100)	803(0·1)	174(14)	174(14)	140(5)	87(27)	86(7)	71(30)
(32)	$C_{37}H_{70}N_2O_{12}$	560(3·5)	561(1·4)	158(100)	158(100)	659(0·8)	174(10)	174(10)	140(3)	87(8)	86(5)	71(23)
(33)	$C_{29}H_{55}NO_{10}$			158(100)			174(31)		140(5)	87(17)	86(8)	71(26)

Compd	Formula	x	y	z	aa	bb	cc	dd	ee	ff''	ll''
(31)	$C_{44}H_{82}N_2O_{15}$	70(9)	98(25)	116(40)	114(9)	100(41)	145(4)	127(9)	109(4)	750(0·3)	793(0·09)
(32)	$C_{37}H_{70}N_2O_{12}$	70(4)	98(13)	116(23)	114(6)	100(27)				606(3·3)	649(0·8)
(33)	$C_{29}H_{55}NO_{10}$	70(7)	98(24)	116(64)							492(20)

TABLE 5

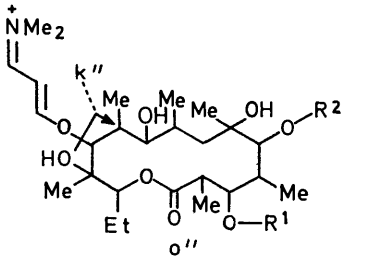
Erythromycin derivatives

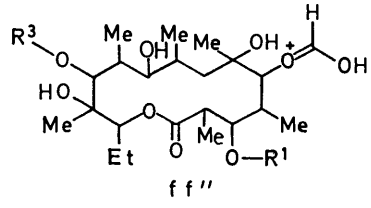
Compound	Formula	M^+	a'''	b'''	c'''	d'''	e'''	f'''	g''	h''	m	q	s
(34)	$C_{37}H_{67}NO_{13}$	733(0·01)	574(0·19)	575(0·07)	558(0·45)	559(0·13)	575(0·07)	576(0·05)	559(0·13)	560(0·05)	158(100)	174(14)	140(5)
(35)	$C_{39}H_{69}NO_{14}$	775(0·02)	616(0·1)	617(0·03)	600(0·1)	601(0·02)					200(43)	216(1·5)	140(24)
(36)	$C_{41}H_{71}NO_{15}$	817(0·03)	616(0·1)	617(0·05)	600(0·3)	601(0·1)	617(0·04)	618(0·02)	601(0·1)	602(0·1)	200(46)	216(4)	140(19)

Compound	Formula	u	v	w	x	y	z	pp	dd	ee	qq	qq and aa'''	qq and bb'''
(34)	$C_{37}H_{67}NO_{13}$	87(19)	86(5)	71(29)	70(4)	98(31)	116(38)	159(27)	127(15)	109(6)	715(1·5)	556(0·53)	557(0·75)
(35)	$C_{39}H_{69}NO_{14}$	129(3) 87(12)	86(6)	71(47)	70(6)	98(48)	116(22)	159(4)	127(11)	109(10)	757(0·2)	598(0·05)	599(0·05)
36)	$C_{41}H_{71}NO_{15}$	129(2) 87(8)	86(5)	71(30)	70(5)	98(38)	116(14)	201(12)	169(3)	109(20)	799(1·5)	598(0·2)	599(0·3)

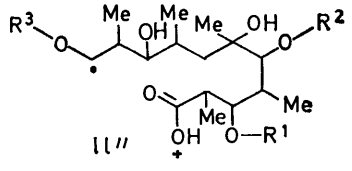
Compound	Formula	qq and cc'''	qq and dd'''	qq and ee'''	qq and ff'''	qq and gg'''	qq and hh'''	rr	ss	tt	kk and/or ll'''	uu
(34)	$C_{37}H_{67}NO_{13}$	540(0·43)	541(0·23)	557(0·75)	558(0·45)	541(0·23)	542(0·11)	269(0·4)	251(0·7)	657(0·26)	648(0·05)	630(0·07)
(35)	$C_{39}H_{69}NO_{14}$	582(0·1)	583(0·08)	557(0·02)	558(0·1)	541(0·1)	542(0·1)	269(0·3)	251(1·0)	699(0·2)		
(36)	$C_{41}H_{71}NO_{15}$	582(0·8)	583(0·4)	599(0·3)	600(0·3)	583(0·4)	584(0·2)	269(0·5)	251(2·1)	741(0·5)		

spectra of the dihydro-series. Losses of 85 mass units from the molecular ions in the dihydro-series gave rise to ions ll'' in all instances.

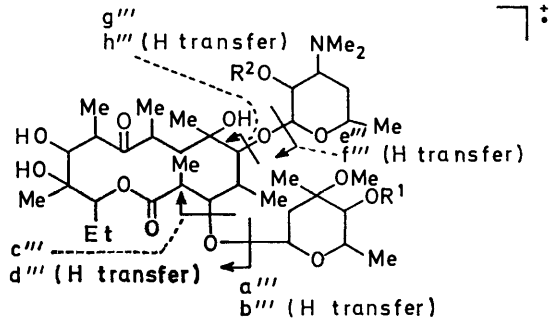




In view of the close similarity between the structures of the megalomicins and erythromycin A (34),[19] the mass

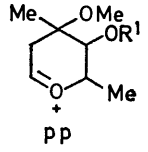


spectra of the latter, and its 2''-*O*-acetyl (35)[20] and 4',2''-di-*O*-acetyl (36) derivatives[20] were recorded (Table 5). In comparison with the megalomicins the erythromycins showed very weak molecular ions. Glycosidic cleavages a'''—d''' of the cladinose unit were apparent in the high mass region, with the accompanying low



SCHEME 13

mass fragment ions pp, dd, and ee. The *m/e* values of the ions pp and dd demonstrated that the methoxy-group was located at C-3' and that the hydroxy-group



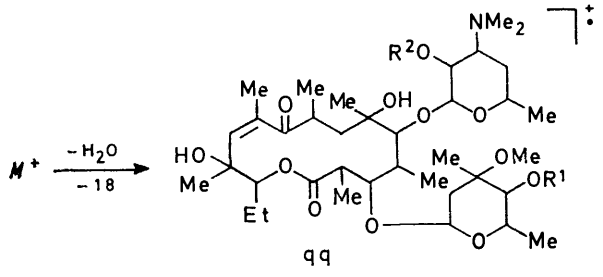
was at C-4' in the erythromycins. In the spectrum of the monoacetate (35) the positions of these ions remained unchanged, indicating that the acetyl group was not in the cladinose unit. In that of the diacetate (36), the *m/e* values of the ions pp and dd had increased by 42

[19] P. F. Wiley, K. Gerzon, E. H. Flynn, M. V. Sigal, O. Weaver, U. C. Quarck, R. C. Chauvette, and R. Monahan, *J. Amer. Chem. Soc.*, 1957, **79**, 6062.

[20] A. Banaszek, J. St. Pyrek, and A. Zamojski, *Roczniki Chemii*, 1969, **43**, 763.

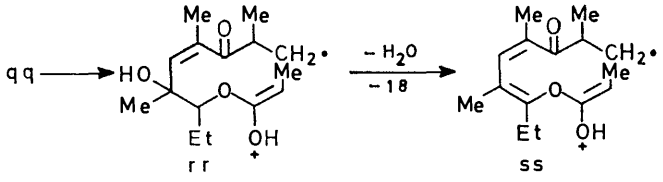
mass units, consistent with the location of one of the acetyl groups at the 4′-position in the cladinose. The positions of the high-mass ions aa′′′—dd′′′ also indicated the presence of acetyl substitution in the cladinose. Glycosidic cleavages of the desosamine also occurred with the erythromycins, leading to fragments e′′′—h′′′, which again varied according to the acylation pattern in that sugar residue. In all cases the base peak in the spectrum was due to the ion m. The shift of the base peak from *m/e* 158 to 200 in the esters (35) and (36) indicated that the hydroxy-group in the desosamine was acetylated in those derivatives. The other low-mass fragment ions from the desosamine, namely q, s, and u—z, were also present in the spectra of the erythromycins.

In contrast to the megalomicins, the erythromicins showed substantial loss of water from the molecular ion, apparently due to the loss of the 11-hydroxy-group to give the ion qq. This loss cannot occur in the megalomicins owing to the presence of the 11-β-D-rhodosaminyloxy-group. Further glycosidic cleavages from the ion



SCHEME 14

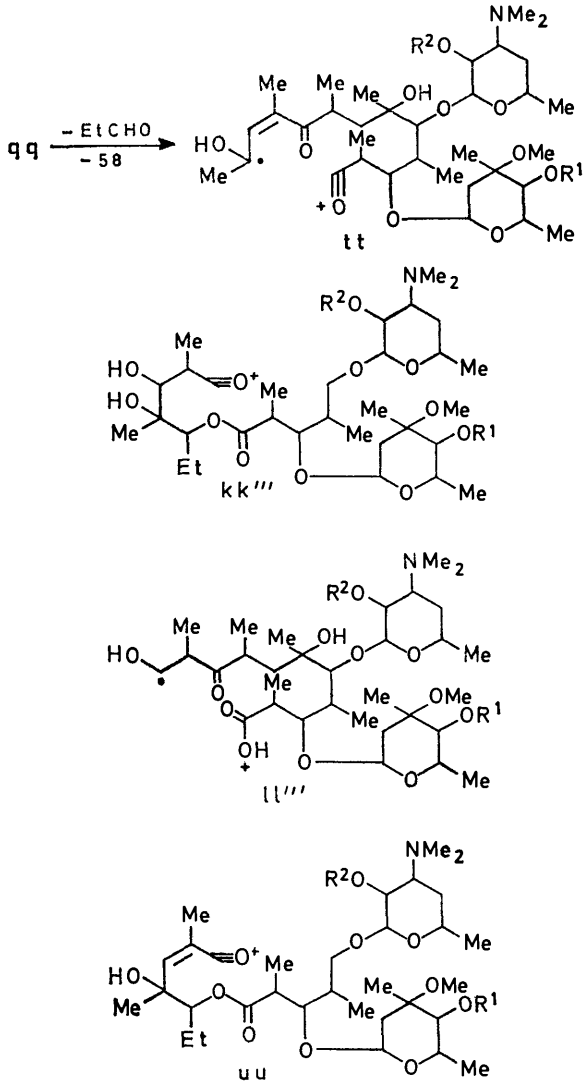
qq by fragmentations of the type a′′′—hh′′′ were also observed in the erythromycins. The fragment qq also underwent a McLafferty rearrangement at the lactone carbonyl group, accompanied by cleavage between C-6 and C-7 to give the ion rr, which underwent a further loss of water to give the fragment ion ss. The formation of the ions rr and ss in the erythromycins is analogous to



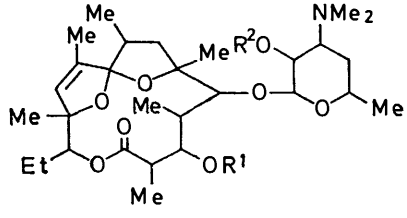
SCHEME 15

the formation of the ions hh and ii in the megalomicins. The loss of propionaldehyde from the ions qq also occurred in the erythromycins to give the ion tt. In the case of erythromycin A (34), a loss of 85 mass units from the molecular ion leading to fragments kk′′′ and ll′′′, as well as a loss of 85 mass units from ion qq leading to ion uu, was observed.

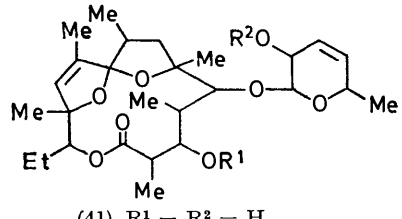
In the course of the degradation of the megalomicins, a number of erythralosamines (37)—(42) [19] were prepared; the mass spectral data for these compounds are



given in Table 6. The glycosidic cleavages e′′′′—h′′′′ of the desosamine unit were observed with the expected

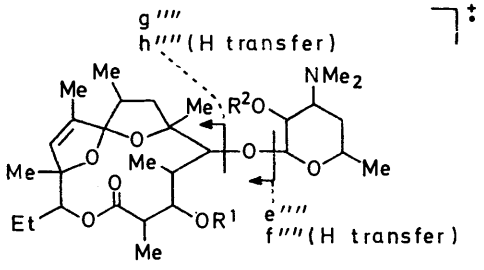


(37) $R^1 = R^2 = H$
(38) $R^1 = H$, $R^2 = Ac$
(39) $R^1 = H$, $R^2 = EtCO$
(40) $R^1 = R^2 = Ac$



(41) $R^1 = R^2 = H$
(42) $R^1 = R^2 = Ac$

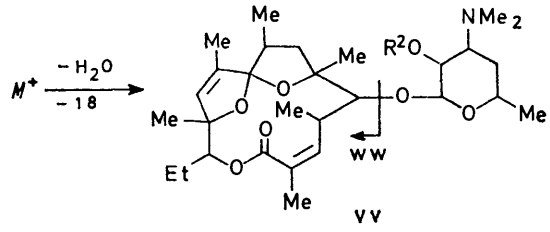
shifts depending on the acylation of the 2′-hydroxy-group, leading to a base peak in all cases due to the ion



SCHEME 16

m. The low mass fragment ions q, s, and u—z from the desosamine were also observed. The mass spectrum of

aglycone of the erythralosamine were also observed. A McLafferty rearrangement at the lactone carbonyl group accompanied by cleavage of the C(5)–C(6) bond gave an



ion xx at *m/e* 295 (Scheme 17) and an ion yy at *m/e* 221 (Scheme 18). A fragment common to all of the ery-

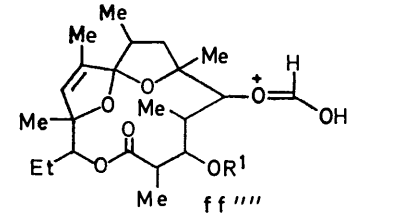
TABLE 6

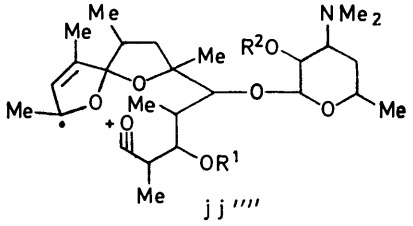
The erythralosamines

Compound	Formula	*M*+	e''''	f''''	g''''	h''''	m	q	s	u	v	w	x
(37)	$C_{28}H_{49}NO_8$	539(3·4)	381(2·9)	382(1)	365(3)	366(0·8)	158(100)	174(11)	140(22)	87(11)	86(3)	71(11)	70(3)
(38)	$C_{31}H_{51}NO_9$	581(0·26)	381(0·59)	382(0·15)	365(0·43)	366(0·11)	200(52)	216(1)	140(8)	129(5) 87(12)	86(6)	71(100)	70(8)
(39)	$C_{32}H_{53}NO_9$	595(1·2)	381(3)	382(0·8)	365(2·4)	366(0·7)	214(100)	230(2·5)	140(23)	143(4) 87(11)	86(5)	71(44)	70(6)
(40)	$C_{33}H_{53}NO_{10}$	623(0·3)	423(2·1)	424(0·6)	407(2·1)	408(0·6)	200(98)	216(1)	140(11)	129(3) 87(7)	86(6)	71(21)	70(5)
(41)	$C_{27}H_{42}O_8$	494(1·5)	381(8)	382(2·5)	365(0·8)	366(0·2)							
(42)	$C_{31}H_{46}O_{10}$	578(1·73)	423(12)	424(3·3)	407(0·6)	408(0·2)							

Compound	Formula	y	z	ff''''	jj''''	vv	ww	xx	yy	zz	Miscellaneous
(37)	$C_{28}H_{49}NO_8$	98(18)	116(18)	411(0·1)	481(2·3)	521(0·7)	347(0·7)	295(0·5)	221(3)	123(29)	
(38)	$C_{31}H_{51}NO_9$	98(24)	116(9)		523(0·09)	563(0·03)	347(0·13)	295(0·19)	221(2)	123(24)	*M* − 60: 521(0·03)
(39)	$C_{32}H_{53}NO_9$	98(47)	116(22)		537(0·4)	577(0·1)	347(0·8)	295(1)	221(4)	123(59)	*M* − 74: 521(0·3)
(40)	$C_{33}H_{53}NO_{10}$	98(80)	116(39)		565(0·4)		347(0·8)		221(4)	123(24)	*M* − 60: 563(0·3)
(41)	$C_{27}H_{42}O_6$				436(0·9)	476(0·4)	347(0·8)	295(1·2)	221(9)	123(89)	nn: 113(27)
(42)	$C_{31}H_{48}O_{10}$				520(1·2)		347(0·5)	295(0·2)	221(13)	123(43)	nn: 113(29) and 155(100)

erythralosamine (37) showed a fragment ion ff'''' formed by the loss of 128 mass units from the molecular ion. As in the case of the megalomicins and megalalosamines, this fragmentation was not observed when the desosamine was acylated. The erythalosamines exhibited a loss of propionaldehyde from the molecular ion giving rise to the ions jj''''. A loss of water from the molecular

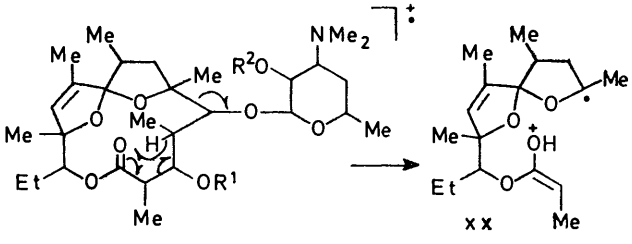




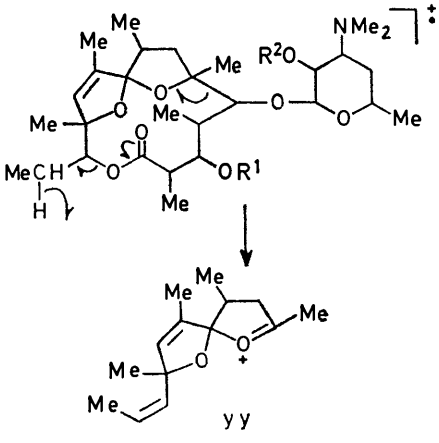
ion to give the ions vv was observed in those erythralosamines where the 3-hydroxy-group was not acylated. Loss of the desosamine and a water molecule from the molecular ion gave rise to the ions ww.

A number of characteristic fragmentations of the

thralosamines at *m/e* 123 is thought to be due to the ion zz (Scheme 19).

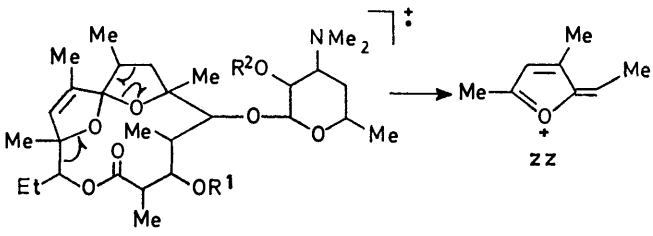


SCHEME 17



SCHEME 18

The deaminated erythralosamine derivatives (41) and (42) showed glycosidic cleavage fragments in the high-mass region similar to those observed with erythralosamine (37), and gave rise to ions nn in the low-mass



SCHEME 19

region. The deaminated erythralosamines (41) and (42) did not lose 128 mass units to give the ions ff'''', further supporting the fact that the latter fragmentation involved loss of a fragment which included the amino-group of the desosamine.

EXPERIMENTAL

Low resolution spectra were run at 70 eV on a Perkin-Elmer RMU-6D spectrometer, or on an Atlas CH5 spectrometer. High resolution spectra were run on an A.E.I. MS902B spectrometer, or a JEOL JMS-01SC spectrometer at 70 eV. Metastable peaks were observed at 10 eV.

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