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Chemical transformation of tylosin derivatives into neutral macrolides having a 3′-methoxyl group

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Abstract

This paper describes the chemical transformation of the basic 16-membered macrolides, tylosin derivatives, into neutral macrolides having a 3′-methoxyl group. 2′,4′-Di-*O*-acetyl-3,23-bis(*O*-*tert*-butyldimethylsilyl)mycaminosyltylonolide 9,20-bis(ethylene acetal) *N*-oxide (**1b**) was treated with Ac_2O–pyridine in CH_2Cl_2 to afford the 3′-ketone **1c** and the 3′-*N*-acetyl-3′-*N*-demethyl derivative **1d** in 67 and 5% yield, respectively. Reduction of **1c** with $Zn(BH_4)_2$ gave the 3′-alcohol **1e** in 84% yield stereoselectively. *O*-Methylation of **1e** with MeOTf and 2,6-di-*tert*-butylpyridine gave the 3′-methyl ether **1f** in 49% yield in spite of the presence of the adjacent acetoxyl groups. Deprotection of **1f** provided the desired neutral macrolide **1g**. Similar synthetic routes were also used for transformation of the suitably protected 4′-deoxymycaminosyltylonolide **2b** and desmycosin **3c** into neutral macrolides having a 3′-methoxyl group. It was found that the mycinose moiety of a neutral macrolide plays an important role in its antimicrobial activity.

Keywords: Sixteen-membered macrolide; Neutral macrolide; Tylosin; The Polonovski reaction; Stereoselective reduction of ketone; *O*-Methylation; Antibiotic

1. Introduction

The 16-membered macrolides may be broadly divided into two classes, namely basic macrolides and neutral macrolides. Tylosin, which has a 3-dimethylamino sugar at the 5-position of the aglycon, belongs to the former class. The latter class is represented by chalcomycin (CM) [1,2] and neutramycin (6-demethylCM) [3] which have a 3-methoxyl group in the 5-*O*-sugar moiety. Recently, two novel neutral 16-membered macrolides, GERI-155 [4] (10,11-dihydroCM) and 250-144C [5] (8-deoxy-10,11-dihydro-12-enoCM), were independently isolated from a fermentation broth of chalcomycin-producing *Streptomyces*. It is noteworthy that the former showed very strong antibacterial activity against *Pseudomonas aeruginosa* and *Salmonella typhimurium*, against which macrolide antibiotics are usually ineffective.

Despite the characteristic difference between these two classes, there exists little difference in their antibacterial spectra. In view of this similarity in microbiological activity, it is considered that

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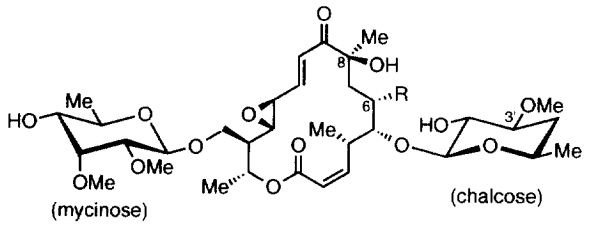
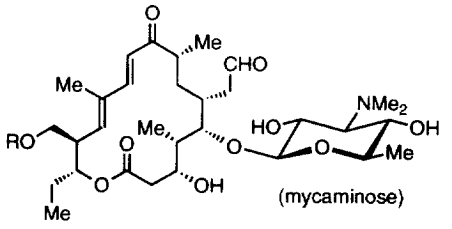
both the dimethylamino and methoxyl groups at the 3′-position participate in like manner in linking to the ribosome and inhibiting protein synthesis in microorganisms [6]. Although several modifications of the 3′-dimethylamino group resulted in diminished antimicrobial activity [7], there were no reports about its replacement with a methoxyl group.



chalcomycin (CM) R = Me
neutramycin R = H



mycaminosyltylonolide (MT) R = H
desmycosin (DM) R = mycinosyl

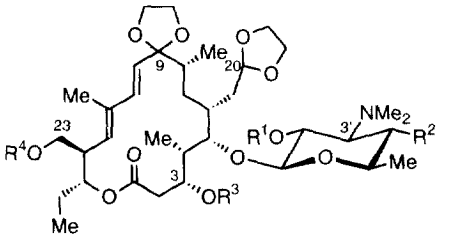
Therefore, our interest was focused on chemical transformation of tylosin derivatives into neutral macrolides having a 3′-methoxyl group in order to investigate the effect of this group on their antimicrobial activity. Our synthetic strategy includes conversion of the 3′-dimethylamino compound into the 3′-ketone by utilizing the Polonovski reaction, stereoselective hydrogenation of the ketone, and *O*-methylation of the resulting alcohol.

The Polonovski reaction [8] has often been utilized to cleave the glycosyl bond between the mycaminose moiety and the aglycon of 16-membered macrolides. For example, treatment of the *N*-oxides of leucomycin A_3, mycaminosyltylonolide (MT) or carbomycin B acetal with Ac_2O–$CHCl_3$ afforded their aglycons [9–11]. The degradation of the trifluoroacetyl derivative of MT *N*-oxide also gave its aglycon [12]. The 3′-keto derivatives and *N*-acetyl-*N*-demethyl derivatives were also isolated from the Polonovski reaction products of leucomycin A_3 [13,14] and mycinamicin I [15]. On the other hand, almost exclusive conversion to the 3′-keto derivative was reported by Girotra and Wendler [16] in the reaction of leucomycin A_3 *N*-oxide with Ac_2O in pyridine. The use of pyridine, however, did not always give a 3′-ketone as the sole product. For instance, Ganguly [17] described that the reaction of rosaramicin *N*-oxide with Ac_2O–pyridine provided a mixture of several compounds. Nicolaou et al. [18] treated MT with trifluoroacetic anhydride–pyridine in CH_2Cl_2 in order to cleave its glycosidic bond.

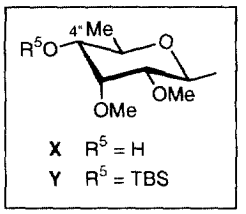
2. Results and discussion

MT [19], obtained by acid hydrolysis of tylosin, afforded compound **1a** [20] via a three-step protection. Treatment of **1a** with 3-chloroperoxybenzoic acid (*m*-CPBA) and $NaHCO_3$ in CH_2Cl_2 gave the *N*-oxide **1b**, which was immediately used for the Polonovski reaction. As a result of extensive trial and error, we have found that the reaction of **1b** with Ac_2O (2 mol equiv) and pyridine (4 mol equiv) in CH_2Cl_2 at room temperature (rt) overnight yielded the 3′-ketone **1c** as the major product. Chromatographic separation allowed the isolation of **1c** and the 3′-*N*-acetyl-3′-*N*-demethyl derivative **1d** in 67 and 5% yields, respectively, from **1a**. The two doublet signals for H-2′ (δ 5.19, J = 8 Hz) and H-4′ (δ 3.62, J = 10 Hz) of **1c** supported the absence of H-3′. The two *N*-methyl signals (δ 2.87 and 2.77 in a 2:1 ratio) for **1d** indicated that it was a mixture of two rotational isomers around C (3′)–N bond in $CDCl_3$. This was proved by the 2D ROESY spectrum of **2i** as described later.

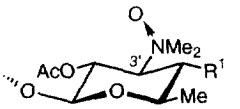
A general mechanism of the Polonovski reaction depicted by Grierson [8] was modified to explain the formation of **1c** and **1d** (Scheme 1). In the first step, reaction of the *N*-oxide **1b** with Ac_2O gives the *O*-acetylimonium acetate **A**. In the second step, the acetate anion participates in the removal of the 3′-proton or the *N*-methyl proton from a carbon atom adjacent to the positively charged nitrogen. Elimination of the 3′-proton (Path a) gives the iminium salt **B**, which is led finally to the 3′-ketone **1c**. This salt **B** is in equilibrium with its addition product **C**. In the third stage, a further attack of Ac_2O on **C** affords the intermediate salt **D**. In the final stage, the acetate anion promotes liberation of Ac_2O and $AcNMe_2$ from **D** to yield **1c**. On the other hand, in the second step, elimination of the *N*-methyl proton (Path b) gives the iminium salt **B′**,



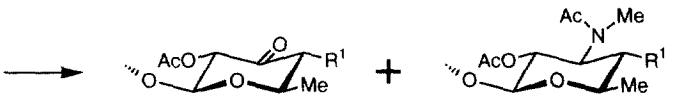
	R^1	R^2	R^3	R^4
1a	Ac	OAc	TBS	TBS
2a	Ac	H	H	H
2b	Ac	H	TBS	TBS
3a	H	OH	H	**X**
3b	Ac	OAc	H	**X**
3c	Ac	OAc	TBS	**Y**



X R^5 = H
Y R^5 = TBS

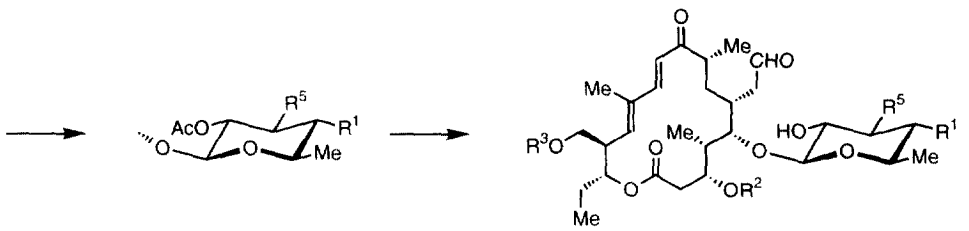


	R^1	R^2	R^3
1b	OAc	TBS	TBS
2c	H	TBS	TBS
3d	OAc	TBS	**Y**



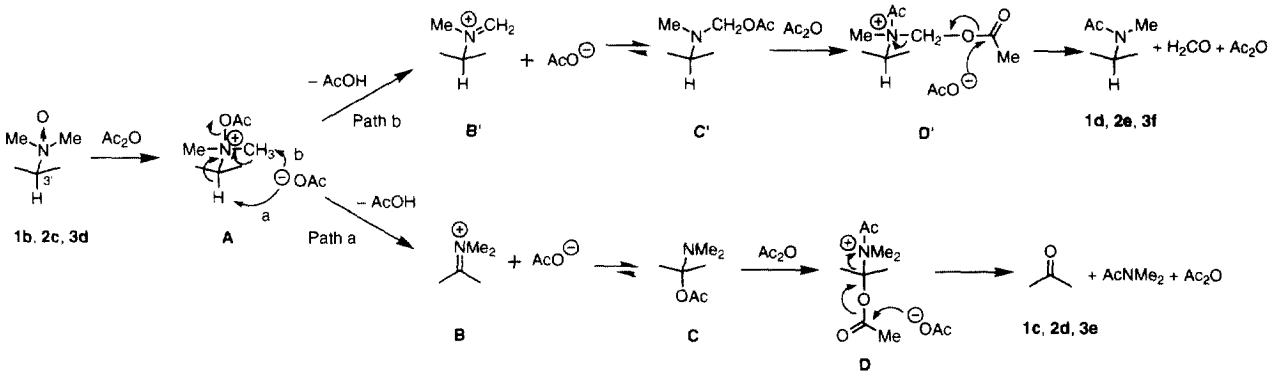
	R^1	R^2	R^3
1c	OAc	TBS	TBS
2d	H	TBS	TBS
3e	OAc	TBS	**Y**

	R^1	R^2	R^3
1d	OAc	TBS	TBS
2e	H	TBS	TBS
3f	OAc	TBS	**Y**



	R^1	R^2	R^3	R^5
1e	OAc	TBS	TBS	OH
1f	OAc	TBS	TBS	OMe
2f	H	TBS	TBS	OH
2g	H	TBS	TBS	OMe
3g	OAc	TBS	**Y**	OH
3h	OAc	TBS	**Y**	OMe

	R^1	R^2	R^3	R^5
1g	OH	H	H	OMe
2h	H	H	H	OMe
2i	H	H	H	NAc(Me)
3i	OH	TBS	**Y**	OMe
3j	OH	H	**X**	OMe



Scheme 1.

which is in equilibrium with the *N*-acetoxymethyl intermediate **C'**. Reaction of **C'** with Ac_2O produces the intermediate salt **D'**, which finally liberates H_2CO and Ac_2O to afford **1d**. From the yields of **1c** and **1d**, it is clear that the reactivity of the 3'-proton of **A** is much higher than that of the *N*-methyl protons. The presence of the two electron-withdrawing acetoxy groups at 2' and 4' nicely accounts for the difference in their kinetic acidity.

Stereoselective reduction of **1c** was performed with $Zn(BH_4)_2$ [21] at −20 °C to give the 3'-hydroxy derivative **1e** in 84% yield. When **1c** was treated with $NaBH_4$ or Bu_4NBH_4, partial deacetylation and migration of the acetyl groups of **1e** were observed because of the reagent's basicity. The stereochemistry of the 3'-hydroxyl group was confirmed by ^1H NMR ($J_{2',3'} = J_{3',4'} = 10$ Hz).

Methylation of **1e** with an excess of MeOTf and 2,6-di-*tert*-butylpyridine (DTBP) [22] in refluxing CH_2Cl_2 for 48 h gave the 3'-*O*-methyl derivative **1f** in 49% yield. The structure of **1f** was supported by the ^1H NMR (3'-OMe, δ 3.45 s). Reactions at higher temperatures in other solvents or conventional methylation (MeI–Ag_2O in DMF) promoted migration of the acetyl groups of **1e** or partial deacetylation to give a complex mixture.

Deacetylation of **1f** under Zemplén conditions (0.1 M methanolic NaOMe, 50 °C, 1.5 h) followed by acidic hydrolysis (1:1 MeCN–0.1 M aq HCl, 50 °C, 9 h) afforded the desired 3'-methoxy analog **1g** of MT in 80% yield. As shown in Table 1, it has become clear that replacement of the 3'-dimethylamino group of MT with a methoxyl group causes total loss of its antimicrobial activity.

Natural neutral macrolides, except for aldgamycins [23] and swalpamycin [24], have chalcose (4-deoxy sugar) as their common sugar at the 5-position. Accordingly, we tried a similar synthetic route, starting from 4'-deoxyMT (DT) [25] to examine the effects of 4'-deoxygenation of **1g**.

Selective acetylation of the 2'-hydroxyl group (Ac_2O, CH_2Cl_2) in DT 9,20-bis(ethylene acetal) [26] furnished **2a** in 73% yield. Silylation of **2a** with *tert*-butyldimethylsilyl trifluoromethanesulfonate (TBSOTf) in the presence of 2,6-lutidine gave the 3,23-disilyl ether **2b** in 79% yield. When TBSCl was used for silylation of **2a**, the 23-monosilyl ether was exclusively obtained. The modified Polonovski reaction of **2b** gave, via **2c**, the 3'-ketone **2d** in 36% yield from **2b** which is lower than that of the 3'-*N*-acetyl-3'-*N*-demethyl derivative **2e** (43% yield from **2b**). This difference in regioselectivity between the *N*-oxides **1b** and **2c** could be ascribed to lower acidity of the 3'-proton of the 4'-deoxy derivative of the intermediate **A** in Scheme 1. The 3'-ketone **2d** was converted stereoselectively into alcohol **2f** [$Zn(BH_4)_2$, 80% yield], then into methyl ether **2g** (44% yield) by methylation (MeOTf, DTBP) of **2f**. Two-step deprotection as described above resulted in the formation of the desired 4'-deoxy-3'-methoxy analog **2h** (58% yield). As shown in Table 1, **2h** showed hardly any antimicrobial activity. Therefore, it is concluded that the 4'-deoxygenation of **1g** has a minimal effect on its antibacterial activity.

Deprotection of **2e** gave the 3'-*N*-acetyl-3'-*N*-demethyl derivative (**2i**) of DT. The ^1H NMR spectra of **2i** indicated that it was a 2:1 mixture of isomers in $CDCl_3$. In the phase-sensitive 2D ROESY spectrum [27,28], the H-3' signals of the major and minor isomers at δ 4.63 and 3.70, respectively, showed a distinct correlation peak

Table 1
Antimicrobial activity (minimum inhibitory concentration (MIC), μg/ml) [a] of **1g**, **2h**, **2i** and **3j** in comparison with MT, DT, DM and CM

Test organism	1g	MT	2h	2i	DT	3j	DM	CM
Staphylococcus aureus 193	>100	1.56	50	>100	0.78	6.25	0.78	0.78
S. aureus 193EMf	>100	>100	>100	>100	>100	>100	6.25	>100
S. aureus FDA209P	>100	0.78	50	>100	0.39	3.12	0.78	0.39
S. aureus MS9861	>100	3.12	>100	>100	0.78	12.5	6.25	1.56
S. aureus MS10225	>100	3.12	>100	>100	0.78	12.5	3.12	0.78
S. aureus Smith	>100	0.78	>100	>100	0.39	3.12	25	0.78
S. aureus MS16526 (MRSA)	>100	>100	>100	>100	>100	12.5	>100	>100
Micrococcus luteus PCI1001	>100	0.78	>100	>100	0.78	0.78	0.39	6.25
Bacillus subtilis NRRL B-558	>100	3.12	>100	>100	1.56	25	1.56	3.12
Corynebacterium bovis 1810	>100	6.25	>100	100	0.78	3.12	1.56	6.25
Escherichia coli NIHJ	>100	12.5	>100	>100	3.12	>100	100	>100
Klebsiella pneumoniae PCI602	>100	1.56	>100	>100	0.78	>100	50	>100

[a] MICs were determined by the agar dilution streak method (2-fold dilution) in 50% aq MeOH with Mueller–Hinton medium (Difco) at 37 °C for 18 h.

having the same phase with diagonal peaks. Although the respective pairs of the H-1′, 2′, and *N*-methyl signals of the two isomers appeared close together, their correlation peaks were also observed in the same phase. This type of signal correlation due to saturation transfer occurs via a conformational exchange process. Thus, it was proved that **2i** was a mixture of two rotational isomers around the C(3′)–N bond. Compound **2i** showed no antimicrobial activity (Table 1).

Next, we tried synthesis of the 3′-methoxy analog having mycinose at the 23-position, which is another common partial structure of neutral 16-membered macrolides. We chose desmycosin (DM) [29], obtained by mild acidic hydrolysis of tylosin, as the starting compound and used a similar synthetic route as mentioned already.

Protection of the 9-ketone and 20-aldehyde groups in DM as a bis(ethylene acetal) **3a** [$CH(OEt)_3$, $CH_2(OH)CH_2(OH)$, CSA, 66% yield] [30], followed by selective acetylation of the 2′ and 4′-hydroxyl groups (Ac_2O, CH_2Cl_2) in **3a** gave **3b** in 84% yield. Silylation of **3b** with TBSOTf furnished the 3,23-disilyl ether **3c** in 79% yield. The modified Polonovski reaction of **3c** gave the 3′-ketone **3e** (64% yield) and the *N*-acetyl-*N*-demethyl derivative **3f** (4% yield). Stereoselective reduction of **3e** with $Zn(BH_4)_2$ afforded the 3′-alcohol **3g** (74% yield), which was then treated with MeOTf and DTBP to give the 3′-methyl ether **3h** (44% yield). Compound **3h** was deprotected under basic and then acidic conditions to afford the desired 3′-methoxy analog (**3j**) of DM (44% yield, in 2 steps).

As shown in Table 1, compound **3j** showed antimicrobial activity with a spectrum similar to that of CM. Therefore, the mycinose moiety, which is a common structure of neutral 16-membered macrolides, is shown to play an important role in their antimicrobial activity. Other biological activity tests of **1g**, **2h**, and **3j** are now under study.

3. Experimental

General methods.—Organic solns were dried over Na_2SO_4 and concd under diminished pressure at or below 40 °C. TLC was performed on precoated Silica Gel 60 F_{254} plates (Art 5715, E. Merck) and detected by spraying the plates with 50% aq H_2SO_4. For column chromatography, Silica Gel 60 (Art 7734, E. Merck) was used. Optical rotations were determined with a Perkin–Elmer 241 polarimeter. FAB-MS spectra were recorded with a Jeol SX-102 spectrometer, using glycerol as the matrix. ^1H NMR spectra were measured at 500 MHz with a Bruker AMX 500 spectrometer for solns in $CDCl_3$ at 30 °C (Tables 2–4). Chemical shifts (δ) were recorded downfield from internal Me_4Si and confirmed by shift-correlated 2D spectra.

*2′,4′-Di-*O*-acetyl-3,23-bis(*O*-tert-butyldimethylsilyl)-3′-de(dimethylamino)-3′-oxomycaminosyltylonolide 9,20-bis(ethylene acetal)* (**1c**) *and 3′-*N*-acetyl-2′,4′-di-*O*-acetyl-3,23-bis(*O*-tert-butyldimethylsilyl)-3′-*N*-demethylmycaminosyltylonolide 9,20-bis(ethylene acetal)* (**1d**).—To a soln of 2′,4′-di-*O*-acetyl-3,23-bis(*O*-*tert*-butyldimethylsilyl)mycaminosyltylonolide 9,20-bis(ethylene acetal) (**1a**, 750 mg, 0.753 mmol) [20] in dry CH_2Cl_2 (15 mL) were added 3-chloroperoxybenzoic acid (*m*-CPBA, 170 mg, 0.985 mmol) and $NaHCO_3$ (85.0 mg, 1.01 mmol). The mixture was stirred at rt for 10 min, after which the TLC (15:1 $CHCl_3$–MeOH, *N*-oxide (**1b**), R_f 0.29; **1a**, R_f 0.78) indicated the reaction was complete. The mixture was washed with satd aq Na_2SO_3 and satd aq $NaHCO_3$, dried, and filtered. To the resulting soln were added Ac_2O (0.142 mL, 1.51 mmol) and dry pyridine (0.244 mL, 3.02 mmol) and the mixture was kept at rt overnight. After washing with satd aq $NaHCO_3$, the organic soln was dried and concd. The residue obtained was submitted to column chromatography (7:1 toluene–EtOAc, then 6:1 toluene–acetone). The fraction having R_f 0.57 (7:1 toluene–acetone; **1b**, R_f 0) gave, on concn, **1c** (490 mg, 67%) as a colorless solid; $[\alpha]_D^{22}$ −54° (*c* 1, $CHCl_3$). Anal. Calcd for $C_{49}H_{83}O_{15}Si_2$: C, 60.78; H, 8.94. Found: C, 61.05; H, 8.72.

The fraction having R_f 0.37 (7:1 toluene–acetone) gave, on concn, **1d** (35.4 mg, 5%) as a colorless solid; $[\alpha]_D^{22}$ −47° (*c* 1, $CHCl_3$). Anal. Calcd for $C_{52}H_{91}NO_{15}Si_2 \cdot 0.5H_2O$: C, 60.32; H, 9.05; N, 1.35. Found: C, 60.28; H, 8.84; N, 1.19.

*2′,4′-Di-*O*-acetyl-3,23-bis(*O*-tert-butyldimethylsilyl)-3′-de(dimethylamino)-3′-hydroxymycaminosyltylonolide 9,20-bis(ethylene acetal)* (**1e**).—To a cooled (−20 °C) soln of **1c** (437 mg, 0.451 mmol) in dry toluene (8.7 mL), 0.4 M $Zn(BH_4)_2$ in tetrahydrofuran (THF, 2 mL, 0.8 mmol) was added and the mixture was kept at −20 °C for 20 min. After addition of 3.5% aq H_2O_2 (5 mL) to the mixture, the organic soln was separated and the aq soln was extracted with $CHCl_3$ (3×3 mL). The organic solns were combined and dried. The residue obtained on concn was purified by column chromatography

Table 2
^{1}H NMR data for 5-*O*-sugar moieties of **1c–1g**, **2a–2i**, and **3a–3j** at 500 MHz in $CDCl_3$

Compound	Chemical shifts[a] (δ, ppm) and multiplicities[b]									
	H-1′	H-2′	H-3′	H-4′ax	H-4′eq	H-5′	H-6′	Ac	Me	OH
1c	4.71 br	5.19 d	—	3.62 d	—	3.45[c]	1.36 d	2.16 s 6H	—	—
1d major	4.58 br	4.86[c]	4.97[c]	4.70 t	—	3.53[c]	1.21 d	2.00, 2.01, 2.02 s	2.87 s *N*-	—
1d minor	4.56[c]	5.03[c]	3.77[c]	4.75[c]	—	3.49[c]	1.24[c]	nd[d]	2.77 s *N*-	—
1e	4.52 br	4.79 dd	3.58[c]	4.67 t	—	3.40 br	1.19 d	2.11 s 6H	—	2.46 d 3′-
1f	4.46 br	4.96 t	3.40[c]	4.82 t	—	3.48 br	1.18 d	2.08, 2.09 s	3.45 s *O*-	—
1g	4.25 d	3.39 ddd	3.09 t	3.17 dt	—	3.30 dq	1.27 d	—	3.65 s *O*-	2.26 d 4′-, 3.50[c] 2′-
2a	4.55 br	4.81 dd	2.73 dt	1.36[c]	1.70[c]	3.49[c]	1.23 d	2.03 s	2.26 s 6H *N*-	—
2b	4.37 br	4.77 t	2.66[c]	1.35[c]	1.67[c]	3.45 br	1.21 d	2.07 s	2.26 s 6H *N*-	—
2d	4.68 br	5.01 d	—	1.56[c]	1.70[c]	3.60[c]	1.34 d	2.17 s	—	—
2e major	4.51 br	4.72 t	4.70[c]	1.52[c]	1.61[c]	3.56[c]	1.21 d	2.01, 2.03 s	2.84 s *N*-	—
2e minor	nd	nd	nd	nd	nd	nd	nd	nd	2.77 s *N*-	—
2f	4.42 br	4.55 dd	3.69[c]	1.45[c]	2.00 ddd	3.47 br	1.21 d	2.12 s	—	nd
2g	4.37 br	4.75 dd	3.32[c]	1.38[c]	2.20[c]	3.45[c]	1.23 d	2.07 s	3.31 s *O*-	—
2h	4.20 d	3.27 t	3.22 dt	1.22[c]	2.04 ddd	3.47[c]	1.19[c]	—	3.42 s *O*-	—
2i major	4.31[e] d	3.37[e] t	4.63 dt	1.47[c]	1.64[c]	3.62 br	1.22[c]	2.11 s	2.91 s *N*-	—
2i minor	4.26[e] d	3.41[e] t	3.70[c]	nd	nd	nd	nd	nd	2.83 s *N*-	—
3a	4.30 d	3.03[c]	2.35 t	4.67 t	—	3.33 dq	1.26 d	—	2.33 s 6H *N*-	—
3b	4.64 br	4.95[c]	2.76 t	4.74 t	—	3.39 dq	1.16 d	2.00, 2.04 s	2.33 s 6H *N*-	—
3c	4.43 br	4.90[c]	2.72 t	4.75 t	—	3.35 dq	1.14 d	2.03, 2.05 s	2.34 s 6H *N*-	—
3e	4.69 br	5.19 d	—	4.90 d	—	3.67[c]	1.36 d	2.15 s 6H	—	—
3f major	4.58[c]	4.73[c]	4.94[c]	4.71 t	—	3.53[c]	1.18 d	1.99, 2.00, 2.01 s	2.87 s *N*-	—
3f minor	4.54[c]	5.01[c]	3.71[c]	4.82[c]	—	3.47[c]	1.17[c]	nd	2.76 s *N*-	—
3g	4.49 br	4.78 dd	3.60[c]	3.40[c]	—	3.40[c]	1.36[c]	2.11 s 6H	—	nd
3h	4.44 br	4.95 dd	3.38[c]	4.81 t	—	3.35[c]	1.17 d	2.08, 2.09 s	3.36 s *O*-	—
3i	4.32 br	3.10 brt	3.19 t	3.30[c]	—	3.72[c]	1.18[c]	—	3.46 s *O*-	—
3j	4.24 d	3.38 dd	3.08 t	3.18[c]	—	3.28[c]	1.26 d	—	3.65 s *O*-	—

Compound	Coupling constants (J, Hz)						
	$J_{1',2'}$	$J_{2',3'}$	$J_{3',4'ax}$	$J_{3',4'eq}$	$J_{4',5'}$	$J_{5',6'}$	Others
1c	8	—	—	—	10	8	—
1d major	nd	nd	10	—	10	6	—
1e	9	10	10	—	10	8	—
1f	10	10	10	—	10	8	—
1g	8	9	9	—	9	6	2 $J_{2',2'\text{-OH}}$, 3 $J_{4',4'\text{-OH}}$
2a	8	10	10	nd	nd	6	—
2b	9	nd	nd	nd	nd	7	—
2d	8	—	—	—	nd	7	—
2e major	8	8	nd	nd	nd	7	—
2f	8	9	nd	4	6 $J_{4'eq,5'}$	6	14 $J_{4'ax,4'eq}$
2g	9	10	nd	nd	nd	8	—
2h	8	9	9	5	6 $J_{4'eq,5'}$	nd	14 $J_{4'ax,4'eq}$
2i major	8	9	10	4	nd	nd	—
2i minor	8	9	nd	nd	nd	nd	—
3a	8	10	10	—	10	6	—
3b	nd	10	10	—	10	8	—
3c	nd	9	9	—	9	6	—
3e	8	—	nd	—	9	6	—
3f major	nd	nd	10	—	10	6	—
3g	9	8	9	—	9	nd	—
3h	8	10	10	—	10	6	—
3i	nd	10	10	—	nd	6	—
3j	8	9	8	—	nd	7	—

[a]Taken from the 1D spectra whenever possible.
[b]s, singlet; d, doublet; t, triplet; q, quartet; br, broad. Multiplicities are not shown for overlapped signals.
[c]Taken from the 2D COSY spectra.
[d]nd: Not determined.
[e]Relative signal intensities are ~2:1 in each column.

(5:1 toluene–acetone) to give **1e** (366 mg, 84%) as a colorless solid; $[\alpha]_D^{21}$ −62° (*c* 1, $CHCl_3$); TLC (6:1 toluene–acetone):R_f 0.32 (**1c**, R_f 0.66). Anal. Calcd for $C_{49}H_{85}O_{15}Si_2$: C, 60.65; H, 8.83. Found: C, 60.50; H, 8.94.

2′,4′-Di-O-acetyl-3,23-bis(O-tert-butyldimethylsilyl)-3′-de(dimethylamino)-3′-methoxymycaminosyltylonolide 9,20-bis(ethylene acetal) (**1f**).—To a soln of **1e** (320 mg, 0.33 mmol) in dry CH_2Cl_2 (6.4 mL) were added 2,6-di-*tert*-butylpyridine (DTBP, 4.14 mL, 18.5 mmol) and MeOTf (1.12 mL, 9.90 mmol) and the mixture was refluxed for 48 h. After addition of MeOH (0.40 mL, 9.89 mmol), the mixture was successively washed with satd aq $NaHCO_3$, satd aq $KHSO_4$, and satd aq $NaHCO_3$, dried and concd. The residue was chromatographed (10:1 toluene–acetone) to afford **1f** (159 mg, 49%) as a colorless solid; $[\alpha]_D^{21}$ −61° (*c* 1, $CHCl_3$); TLC (10:1 toluene–acetone):R_f 0.38 (**1e**, R_f 0.15). Anal. Calcd for $C_{50}H_{87}O_{15}Si_2$: C, 61.00; H, 8.91. Found: C, 61.14; H, 8.93.

3′-De(dimethylamino)-3′-methoxymycaminosyltylonolide (**1g**).—A soln of **1f** (85.0 mg, 0.086 mmol) in 0.1 M methanolic NaOMe (2 mL) was heated at 50 °C for 1.5 h. After cooling the soln was neutralized with Dowex 50W-X8 resin (H^+ type), filtered, and concd to give the deacetylated compound (76.6 mg, 99%) as a colorless solid; TLC (4:1 toluene–acetone):R_f 0.38 (**1f**, R_f 0.67). The solid was dissolved in MeCN (0.8 mL), 0.1 M aq HCl (0.8 mL) was added, and the mixture was heated at 50 °C for 9 h. After neutralization with Dowex 1X2 resin (OH^- type), the soln was filtered, dried, and concd. The residue was chromatographed (10:1:0.1 $CHCl_3$–MeOH–28% aq NH_3) to yield **1g** (39.8 mg, 80%) as a colorless solid; $[\alpha]_D^{22}$ −16° (*c* 1, MeOH); TLC (10:1:0.1 $CHCl_3$–MeOH–28% aq NH_3):R_f 0.20, FAB-MS: *m/z* 585 $(M+1)^+$. Anal. Calcd for $C_{30}H_{48}O_{11}\cdot H_2O$: C, 59.78; H, 8.36. Found: C, 59.84; H, 8.32.

2′-O-Acetyl-4′-deoxymycaminosyltylonolide 9,20-bis(ethylene acetal) (**2a**).—To a soln of 4′-deoxymycaminosyltylonolide 9,20-bis(ethylene acetal) (870 mg, 1.30 mmol) [26] in dry CH_2Cl_2 (18 mL) was added Ac_2O (0.16 mL, 1.70 mmol) and the mixture was kept at rt for 2 h. The reaction mixture was washed with satd aq $NaHCO_3$ (3×5 mL), dried, and concd. The residue obtained was chromatographed (1:1 toluene–acetone) to afford **2a** (670 mg, 73%) as a colorless solid; $[\alpha]_D^{24}$ −3° (*c* 1, $CHCl_3$); TLC (1:1 toluene–acetone):R_f 0.40 (starting

Table 3
^{1}H NMR data for 23-*O*-mycinosyl moieties of **3a**—**3j** at 500 MHz in $CDCl_3$

Compound	Chemical shifts[a] (δ, ppm) and multiplicities[b]										
	H-1″	H-2″	H-3″	H-4″	H-5″	H-6″	2″-*O*-Me	3″-*O*-Me	*t*-Bu	Me_2Si	4″-OH
3a	4.55 d	3.03 dd	3.76 t	3.19 dd	3.51[c]	1.26 d	3.49 s	3.62 s	—	—	nd[d]
3b	4.55 d	3.03 dd	3.75 t	3.18 dt	3.51[c]	1.26 d	3.49 s	3.61 s	—	—	2.28 d
3c	4.57 d	2.93 dd	3.62 t	3.28 dd	3.76[c]	1.18 d	3.44 s	3.59 s	0.92 s	0.09, 0.12 s	—
3e	4.58 d	2.94 dd	3.63 t	3.28 dd	3.76 dq	1.18 d	3.45 s	3.59 s	0.92 s	0.09, 0.12 s	—
3f	4.57 d	2.94 dd	3.62[c]	3.28 dd	3.76[c]	1.17 d	3.44 s	3.59 s	0.90 s	0.09, 0.12 s	—
3g	4.57 d	2.94 dd	3.63 t	3.28 dd	3.76 dq	1.18 d	3.44 s	3.59 s	0.92 s	0.09, 0.11 s	—
3h	4.57 d	2.93 dd	3.62 t	3.28 dd	3.75 dq	1.18 d	3.44 s	3.59 s	0.92 s	0.09, 0.12 s	—
3i	4.58 d	2.94 dd	3.63 t	3.28 dd	3.76 dq	1.18 d	3.46 s	3.59 s	0.92 s	0.09, 0.12 s	—
3j	4.56 d	3.03 dd	3.75 t	3.18[c]	3.52[c]	1.26 d	3.49 s	3.62 s	—	—	2.32 d

Compound	Coupling constants (*J*, Hz)					
	$J_{1'',2''}$	$J_{2'',3''}$	$J_{3'',4''}$	$J_{4'',5''}$	$J_{5'',6''}$	$J_{4'',4''\text{-OH}}$
3a	8	4	4	10	6	0
3b	8	3	3	10	6	10
3c	8	4	4	10	7	—
3e	8	3	3	9	7	—
3f	8	3	3	10	7	—
3g	8	3	3	9	6	—
3h	8	3	3	9	6	—
3i	8	4	4	10	6	—
3j	8	4	4	nd	7	10

[a]Taken from the 1D spectra whenever possible.
[b]s, singlet; d, doublet; t, triplet; q, quartet; br, broad. Multiplicities are not shown for overlapped signals.
[c]Taken from the 2D COSY spectra.
[d]nd: Not determined.

Table 4
^{1}H NMR data for aglycons of **1c–1g**, **2a–2i**, and **3a–3j** at 500 MHz in $CDCl_3$

Proton	Chemical shifts[a] (δ, ppm) and multiplicities[b]																					
	1c	**1d**	**1e**	**1f**	**1g**	**2a**	**2b**	**2d**	**2e**	**2f**	**2g**	**2h**	**2i**	**3a**	**3b**	**3c**	**3e**	**3f**	**3g**	**3h**	**3i**	**3j**
H-2a	2.19 br	2.13 br	2.21 d	2.21 d	2.21 d	1.94[c]	2.19 brd	2.30[c]	2.22 br	2.21 br	2.18 brd	1.95 d	1.92[c]	1.94 d	1.93 d	2.22 d	2.14[c]	2.17 br	2.18[c]	2.15 br	2.22 d	1.94 d
H-2b	2.41 dd	2.41 dd	2.41 dd	2.41 dd	2.52 dd	2.44 dd	2.40 dd	2.40 dd	2.40 br	2.40 dd	2.40 dd	2.40 dd	2.49 dd	2.42 dd	2.41 dd	2.40 dd	2.41 dd	2.42 br	2.41 dd	2.40 dd	2.43 dd	2.50 dd
H-3	4.02 br	4.01 br	4.02 br	4.01 br	3.85 d	3.63[c]	4.02 br	4.02 br	4.02 br	4.03 br	4.01 br	3.86 d	3.85 d	3.60[c]	nd[d]	4.00 br	4.01 br	4.01 br	4.01 br	3.99 br	4.05 br	3.84 br
H-4	1.60[c]	1.45 br	1.45 br	1.44 br	1.66[c]	1.46[c]	1.45[c]	1.49 dq	1.44 dq	1.45[c]	1.43[c]	1.71[c]	1.63[c]	1.48 br	nd	1.43 dq	1.49 dq	1.44 dq	1.44 br	1.45 dq	1.47[c]	1.60[c]
H-5	3.75[c]	3.71[c]	3.55[c]	nd	3.73[c]	3.83[c]	nd	nd	nd	nd	3.66[c]	3.75[c]	3.72[c]	3.78[c]	3.80[c]	3.67[c]	3.81[c]	3.75[c]	3.69[c]	3.67[c]	nd	3.65[c]
H-6	2.16[c]	2.01[c]	2.16[c]	nd	2.16[c]	2.21[c]	nd	nd	nd	nd	nd	2.11 br	2.13 br	2.26 d	2.20 d	nd	nd	nd	nd	nd	nd	2.14 br
H-7a	nd	nd	nd	nd	1.49[c]	1.20[c]	1.20[c]	nd	nd	1.35[c]	1.20[c]	1.55[c]	1.54[c]	1.30[c]	1.21[c]	1.26[c]	1.20[c]	nd	nd	1.02[c]	nd	1.48 m
H-7b	nd	nd	nd	nd	1.63[c]	1.51[c]	1.35[c]	nd	nd	1.40[c]	1.40[c]	1.60[c]	1.64[c]	1.58[c]	1.50[c]	1.40[c]	1.31[c]	nd	nd	1.21[c]	nd	1.63[c]
H-8	nd	1.86[c]	1.89[c]	1.89[c]	2.62[c]	1.93[c]	1.50[c]	1.92 br	1.83[c]	1.89 br	1.89 br	2.59 br	2.65 br	1.87[c]	1.89[c]	1.88 m	1.91 br	1.90[c]	1.88[c]	1.89 m	1.86 m	2.59 br
H-10	5.55 d	5.54 d	5.55 d	5.55 d	6.30 d	5.70 d	5.55 d	5.56 d	5.54 d	5.55 d	5.54 d	6.31 d	6.29 d	5.68 d	5.63 d	5.51 d	5.52 d	5.51 d	5.52 d	5.51 d	5.53 d	6.27 d
H-11	6.34 d	6.34 d	6.33 d	6.34 d	7.32 d	6.40 d	6.35 d	6.34 d	6.34 d	6.34 d	6.34 d	7.33 d	7.30 d	6.33 d	6.36 d	6.33 d	6.33 d	6.33 d	6.32 d	6.32 d	6.32 d	7.33 d
H-13	5.38 d	5.37 d	5.38 d	5.36 d	5.84 d	5.33 d	5.36 d	5.38 d	5.37 d	5.37 d	5.36 d	5.85 d	5.83 d	5.42 d	5.41 d	5.40 d	5.41 d	5.41 d	5.41 d	5.40 d	5.43 d	5.92 d
H-14	2.67 ddt	2.67 br	2.67 ddt	2.67 ddt	2.90 ddt	2.84 m	2.68[c]	2.67 ddt	2.67 br	2.67 m	2.67 m	2.89 m	2.90[c]	2.87 ddt	2.87 ddt	2.83 br	2.83 br	2.83 br	2.83 br	2.83 br	2.84 br	2.95[c]
H-15	4.91 dt	4.91[c]	4.91 dt	4.91 dt	4.95 dt	4.88 dt	4.91 dt	4.90 dt	4.89 dt	4.91 dt	4.91 dt	4.94 dt	4.95 dt	4.98 dt	4.96[c]	4.88[c]	4.89 dt	4.90[c]	4.89 dt	4.89 dt	4.90 br	4.99 dt
H-16a	1.52[c]	1.55[c]	1.52[c]	1.55[c]	1.63[c]	1.60[c]	1.56[c]	1.58[c]	1.58[c]	1.51[c]	1.56[c]	1.61[c]	1.54[c]	1.53[c]	1.60 ddq	1.57[c]	1.57[c]	1.56[c]	1.56[c]	1.58[c]	1.53[c]	1.60[c]
H-16b	1.86[c]	1.86 ddq	1.86 ddq	1.86 ddq	1.86 ddq	1.79[c]	1.80 ddq	1.86 ddq	1.86 ddq	1.85 ddq	1.85 ddq	1.85 ddq	1.85[c]	1.88[c]	1.88 ddq	1.84 ddq	1.84 ddq	1.89[c]	1.84 ddq	1.73[c]	1.81[c]	1.81[c]
H-17	0.90[c]	0.89[c]	0.92 t	0.94 t	0.95 t	0.94 t	0.90[c]	0.91 t	0.89[c]	0.92 t	0.92 t	0.95 t	0.95 t	0.93 t	0.93 t	0.93[c]	0.92[c]	0.92[c]	0.92[c]	0.99[c]	0.92[c]	0.94 t
H-18	0.85[c]	0.85[c]	0.84[c]	0.84[c]	1.02[c]	0.82 br	0.85[c]	0.86[c]	0.86[c]	0.87[c]	0.87 d	1.04 d	1.02 d	0.91 d	0.79 br	0.84 d	0.84[c]	0.86 d	0.85 d	0.83 d	0.91[c]	1.01 d
H-19a	1.75[c]	1.68[c]	1.54[c]	1.65[c]	2.41 brd	2.03[c]	1.59[c]	1.72[c]	1.65[c]	1.64[c]	1.61[c]	2.43 dd	2.38 dd	1.23[c]	2.00[c]	1.75[c]	1.79[c]	1.70[c]	1.67[c]	1.58[c]	nd	2.40[c]
H-19b	2.05[c]	2.00[c]	2.00[c]	2.00[c]	2.93 ddd	2.17[c]	2.05[c]	2.08 br	2.01 br	2.05 br	2.05[c]	2.99 ddd	2.92[c]	2.11 ddd	2.11[c]	1.99[c]	2.04 br	1.99[c]	1.97[c]	1.97[c]	nd	2.95[c]
H-20	5.00[c]	4.99[c]	4.99[c]	4.99 br	9.70 s	4.98 br	5.06 br	5.03 br	5.04 br	5.04 br	5.05 br	5.05 br	9.71 s	4.94 br	4.97[c]	5.01[c]	5.00[c]	5.00[c]	4.99[c]	5.00 br	4.99 br	9.69 s
H-21	1.06 d	1.04 d	1.04 d	1.03 d	1.22 d	1.02 d	1.04 d	1.05 d	1.04 d	1.04 d	1.03 d	1.22[c]	1.21[c]	0.99 d	1.00 d	1.03 d	1.05 d	1.03 d	1.03 d	1.03 d	1.03 d	1.21 d
H-22	1.71 s	1.74 s	1.71 s	1.71 s	1.83 s	1.75 s	1.71 s	1.71 s	1.70 s	1.71 s	1.71 s	1.82 s	1.81 s	1.73 s	1.69 s	1.67 s	1.68 s	1.68 s	1.68 s	1.68 s	1.68 s	1.79 s
H-23a	3.62[c]	3.62[c]	3.62[c]	3.62[c]	3.73[c]	3.50 dd	3.61[c]	3.62[c]	3.59[c]	3.62[c]	3.60[c]	3.73[c]	3.73[c]	3.49[c]	3.49[c]	3.41[c]	3.41[c]	3.41[c]	3.41[c]	3.41[c]	3.41[c]	3.53[c]
H-23b	3.62[c]	3.62[c]	3.62[c]	3.62[c]	3.73[c]	3.72 dd	3.61[c]	3.62[c]	3.59[c]	3.62[c]	3.60[c]	3.73[c]	3.73[c]	3.95[c]	3.94[c]	3.96[c]	3.95[c]	3.95[c]	3.94[c]	3.94[c]	3.95[c]	3.99[c]
t-Bu	0.84 s,	0.87 s,	0.87 s,	0.87 s,	—	—	0.87 s,	0.84 s,	0.87 s,	0.86 s,	0.87 s,	—	—	—	—	0.88 s	0.84 s	0.88 s	0.84 s	0.88 s	0.86 s	—
	0.88 s	0.89 s	0.88 s	0.88 s			0.88 s	0.87 s	0.88 s	0.87 s	0.88 s											
Me_2Si	0.007 s,	0.00 s,	0.01 s,	0.01 s,		—	0.003 s,	0.003 s,	0.002 s,	0.001 s,	0.004 s,	—	—	—	—	0.01 s,	0.01 s,	0.00 s,	0.001 s,	0.02 s,	0.09 s,	—
	0.011 s,	0.01 s,	0.02 s,	0.02 s			0.01 s,	0.01 s,	0.01 s,	0.01 s,	0.01 s,					0.14 s	0.14 s	0.13 s	0.14 s	0.16 s	0.12 s	
	0.02 s,	0.02 s,	0.03 s,	0.03 s,			0.02 s,	0.02 s,	0.02 s,	0.02 s,	0.02 s,											
	0.07 s	0.15 s	0.04 s	0.04			0.03 s	0.10 s	0.11 s	0.03 s	0.03 s											

[a]Taken from the 1D spectra whenever possible. Signals for ethylene-acetal protons appear as mutiplet at δ 3.5–4.0 for all compounds except for **7**, **15**, **16**, and **26**.
[b]s, singlet; d, doublet; t, triplet; q, quartet; m, multiplet; br, broad. Multiplicities are not shown for overlapped signals.
[c]Taken from the 2D COSY spectra.
[d]nd: Not determined.

Although the coupling constants for the aglycons are not always detectable due to signal overlapping, the range of their values are as follows. 2a,2b = 16–19 Hz; 2b,3 = 6–12 Hz; 3,4 = 3 Hz; 4,18 = 6–8 Hz; 6,19b = 9–10 Hz; 7a,7b = 15 Hz; 7a,8 = 3 Hz; 8,21 = 6–8 Hz; 10,11 = 16–17 Hz; 13,14 = 10–12; 14,15 = 9–12 Hz; 14,23 = 4–5 Hz; 15,16a = 9–12 Hz; 15,16b = 2–4 Hz; 16a,16b = 14–16 Hz; 16a,17 = 6–9 Hz; 16b,17 = 6–9 Hz; 19a,19b = 14–19 Hz; 19a,20 = 3 Hz; 19b,20 = 1–3 Hz.

compound, R_f 0). Anal. Calcd for $C_{37}H_{61}NO_{12}\cdot 0.5\ H_2O$: C, 61.65; H, 8.64; N, 1.97. Found: C, 61.53; H, 8.68; N, 2.04.

2'-O-Acetyl-3,23-bis(O-tert-butyldimethylsilyl)-4'-deoxymycaminosyltylonolide 9,20-bis(ethylene acetal) (**2b**).—To an ice-cold M soln of **2a** (670 mg, 0.94 mmol) in CH_2Cl_2 (0.94 mL) were added 2,6-lutidine (0.59 mL, 5.07 mmol) and *tert*-butyldimethylsilyl trifluoromethanesulfonate (TBSOTf, 0.58 mL, 2.52 mmol) and the mixture was kept at 0 °C for 20 min. After addition of MeOH (0.1 mL, 2.47 mmol) the mixture was concd. The residue obtained was dissolved in $CHCl_3$ (35 mL) and the soln was successively washed with satd aq $NaHCO_3$, satd aq $KHSO_4$, and satd aq $NaHCO_3$, dried and concd. The residue was chromatographed (4:1 toluene–acetone) to afford **2b** (698 mg, 79%) as a colorless solid; $[\alpha]_D^{23}$ −52° (*c* 1, $CHCl_3$); TLC (4:1 toluene–acetone):R_f 0.32 (2'-acetate, R_f 0). Anal. Calcd for $C_{49}H_{89}NO_{12}Si_2$: C, 62.58; H, 9.54; N, 1.49. Found: C, 62.50; H, 9.59; N, 1.54.

2'-O-Acetyl-3,23-bis(O-tert-butyldimethylsilyl)-3'-de(dimethylamino)-4'-deoxy-3'-oxomycaminosyltylonolide 9,20-bis(ethylene acetal) (**2d**) *and 3'-N-acetyl-2'-O-acetyl-3,23-bis(O-tert-butyldimethylsilyl)-3'-N-demethyl-4'-deoxymycaminosyltylonolide 9,20-bis(ethylene acetal)* (**2e**).—To a soln of **2b** (697 mg, 0.741 mmol) in dry CH_2Cl_2 (14 mL) were added *m*-CPBA (167 mg, 0.968 mmol) and $NaHCO_3$ (81 mg, 0.964 mmol) and the mixture was stirred at rt for 10 min. Work-up as described for preparation of **1b** gave a soln of *N*-oxide (**2c**); TLC (15:1 $CHCl_3$–MeOH): R_f 0.22 (**2b**, R_f 0.45). To the soln were added Ac_2O (0.176 mL, 1.87 mmol) and dry pyridine (0.302 mL, 3.73 mmol) and the mixture was kept at rt overnight. Work-up as described for **1c** and **1d** gave a crude mixture of **2d** and **2e**, which were separated by column chromatography (7:1 toluene–EtOAc, and then 6:1 toluene–acetone). Concn of the fraction having R_f 0.67 (6:1 toluene–acetone) gave **2d** (242 mg, 36%) as a colorless solid; $[\alpha]_D^{23}$ −79° (*c* 1, $CHCl_3$). Anal. Calcd for $C_{47}H_{82}O_{13}Si_2$: C, 61.92; H, 9.07. Found: C, 62.00; H, 9.09.

Concn of the fraction having R_f 0.45 (6:1 toluene–acetone) gave **2e** (309 mg, 43%) as a colorless solid; $[\alpha]_D^{23}$ −64° (*c* 1, $CHCl_3$). Anal. Calcd for $C_{50}H_{89}NO_{13}Si_2$: C, 62.01; H, 9.26; N, 1.45. Found: C, 61.95; H, 9.39; N, 1.54.

2'-O-Acetyl-3,23-bis(O-tert-butyldimethylsilyl)-3'-de(dimethylamino)-4'-deoxy-3'-hydroxymycaminosyltylonolide 9,20-bis(ethylene acetal) (**2f**).—To a cooled (−20 °C) soln of **2d** (242 mg, 0.265 mmol) in dry toluene (5 mL), 0.4 M $Zn(BH_4)_2$ in THF (1.5 mL, 0.6 mmol) was added and the mixture was kept at −20 °C for 20 min. The residue obtained after work-up as described above for **1e** was purified by column chromatography (5:1 toluene–acetone) to give **2f** (194 mg, 80%) as a colorless solid; $[\alpha]_D^{23}$ −68° (*c* 1, $CHCl_3$); TLC (4:1 toluene–acetone):R_f 0.38 (**2d**, R_f 0.70). Anal. Calcd for $C_{47}H_{84}O_{13}Si_2$: C, 61.81; H, 9.27. Found: C, 61.92; H, 9.29.

2'-O-Acetyl-3,23-bis(O-tert-butyldimethylsilyl)-3'-de(dimethylamino)-4'-deoxy-3'-methoxymycaminosyltylonolide 9,20-bis(ethylene acetal) (**2g**).—To a soln of **2f** (192 mg, 0.211 mmol) in dry CH_2Cl_2 (5 mL) were added DTBP (1.9 mL, 8.47 mmol) and MeOTf (0.48 mL, 4.24 mmol) and the mixture was refluxed for 48 h. After addition of MeOH (0.17 mL, 4.20 mmol), the mixture was worked up as described above for preparation of **1f**. The residue was chromatographed (7:1 toluene–acetone) to afford **2g** (86.4 mg, 44%) as a colorless solid; $[\alpha]_D^{23}$ −45° (*c* 1, $CHCl_3$); TLC (6:1 toluene–acetone):R_f 0.52 (**2f**, R_f 0.25). Anal. Calcd for $C_{48}H_{86}O_{13}Si_2$: C, 62.17; H, 9.35. Found: C, 62.06; H, 9.29.

3'-De(dimethylamino)-4'-deoxy-3'-methoxymycaminosyltylonolide (**2h**).—A soln of **2g** (80.0 mg, 0.086 mmol) in 0.1 M methanolic NaOMe (1.6 mL) was heated at 50 °C for 3 h. Work-up as described above for **1g** gave the deacetylated compound (75.8 mg, 99%) as a colorless solid; TLC (6:1 toluene–acetone):R_f 0.37 (**2g**, R_f 0.52). The solid was dissolved in MeCN (1.5 mL), 0.1 M aq HCl (1.5 mL) was added, and the mixture was heated at 50 °C for 9 h. After work-up as described above, purification by chromatography (12:1:0.1 $CHCl_3$–MeOH–28% aq NH_3) gave **2h** (28.3 mg, 58%) as a colorless solid; $[\alpha]_D^{22}$ −15° (*c* 1, $CHCl_3$); TLC (12:1:0.1 $CHCl_3$–MeOH–28% aq NH_3): R_f 0.25; FAB-MS: *m/z* 569 $(M+1)^+$. Anal. Calcd for $C_{30}H_{48}O_{10}\cdot 0.75H_2O$: C, 61.89; H, 8.57. Found: C, 61.68; H, 8.27.

3'-N-Acetyl-3'-N-demethyl-4'-deoxymycaminosyltylonolide (**2i**).—Compound **2e** (145 mg, 0.15 mmol) was treated in 0.1 M methanolic NaOMe (3 mL) at rt for 6 h. The deacetylated derivative (138 mg) obtained was then processed with 0.1 M HCl (2.5 mL) in MeCN (2.5 mL) at 50 °C for 9 h. Work-up as described for **1g** followed by chromatography (10:1:0.1 $CHCl_3$–MeOH–28% aq NH_3) gave **2i** (38.9 mg, 42%) as a

colorless solid; $[\alpha]_D^{22}$ −8° (*c* 1, $CHCl_3$); TLC (10:1:0.1 $CHCl_3$–MeOH–28% aq NH_3): R_f 0.37; the designation of peaks having the same phase with diagonal peaks in phase-sensitive 2D ROESY spectra (spin-locking time, 200 msec): H-1′(major),H-1′(minor); H-2′(major),H-2′(minor); H-3′(major),H-3′(minor); *N*-Me(major),*N*-Me(minor); FAB-MS: *m/z* 611 $(M+1)^+$. Anal. Calcd for $C_{32}H_{51}NO_{10}\cdot 1.5H_2O$: C, 60.36; H, 8.55; N, 2.20. Found: C, 60.27; H, 8.43; N, 1.91.

Desmycosin 9,20-bis(ethylene acetal) (**3a**).—To a soln of desmycosin (2.50 g, 3.18 mmol) [29] in 5:1 dry toluene–dry MeCN (6 mL) were added triethyl orthoformate (7.9 mL, 47.5 mmol), dry ethylene glycol (5.4 mL, 96.8 mmol), and camphorsulfonic acid (1.22 g, 5.25 mmol). After 5 h at rt, the mixture was poured into satd aq $NaHCO_3$ (22 mL) with stirring. The organic phase was separated, the aq phase was washed with $CHCl_3$ (3×5 mL), and combined organic solns were dried and concd. The residue obtained was purified by chromatography (10:1:0.1 $CHCl_3$–MeOH–28% aq NH_3) to give **3a** (1.83 g, 66%) as a colorless solid; $[\alpha]_D^{22}$ −25° (*c* 1, $CHCl_3$), lit. +20° [30]; TLC (10:1:0.1 $CHCl_3$–MeOH–28% aq NH_3): R_f 0.42 (desmycosin, R_f 0.32). Anal. Calcd for $C_{43}H_{73}NO_{16}\cdot H_2O$: C, 58.82; H, 8.61; N, 1.60. Found: C, 58.77; H, 8.35; N, 1.73.

2′,4′-Di-O-acetyldesmycosin 9,20-bis(ethylene acetal) (**3b**).—To a soln of **3a** (1.73 g, 1.98 mmol) in dry CH_2Cl_2 (35 mL) was added Ac_2O (0.47 mL, 4.98 mmol) and the mixture was kept at rt for 4 h. The residue obtained after work-up as described for **2a** was chromatographed (3:1 toluene–acetone) to give **3b** (1.59 g, 84%) as a colorless solid; $[\alpha]_D^{23}$ −45° (*c* 1, $CHCl_3$); TLC (3:1 toluene–acetone): R_f 0.32 (**3a**, R_f 0.32). Anal. Calcd for $C_{47}H_{77}NO_{18}$: C, 59.79; H, 8.22; N, 1.48. Found: C, 59.76; H, 8.03; N, 1.44.

2′,4′-Di-O-acetyl-3,4″-bis(O-tert-butyldimethylsilyl)desmycosin 9,20-bis(ethylene acetal) (**3c**).—To an ice-cold M soln of **3b** (1.57 g, 1.66 mmol) in CH_2Cl_2 (1.66 mL) were added 2,6-lutidine (1.23 mL, 10.6 mmol) and TBSOTf (1.22 mL, 5.31 mmol) and the mixture was kept at 0 °C for 20 min. Work-up as described above for **2b** gave a crude solid, which was chromatographed (8:1 toluene–acetone) to afford **3c** (1.55 g, 79%) as a colorless solid; $[\alpha]_D^{22}$ −57° (*c* 1, $CHCl_3$); TLC (8:1 toluene–acetone): R_f 0.37 (**3b**, R_f 0). Anal. Calcd for $C_{59}H_{105}NO_{18}Si_2$: C, 60.43; H, 9.02; N, 1.19. Found: C, 60.55; H, 9.20; N, 1.15.

2′,4′-Di-O-acetyl-3,4″-bis(O-tert-butyldimethylsilyl)-3′-de(dimethylamino)-3′-oxodesmycosin 9,20-bis(ethylene acetal) (**3e**) *and 3′-N-acetyl-2′,4′-di-O-acetyl-3,4″-bis(O-tert-butyldimethylsilyl)-3′-N-demethyldesmycosin 9,20-bis(ethylene acetal)* (**3f**).—To a soln of **3c** (751 mg, 0.64 mmol) in dry CH_2Cl_2 (15 mL) were added *m*-CPBA (154 mg, 0.892 mmol) and $NaHCO_3$ (75 mg, 0.893 mmol) and the mixture was stirred at rt for 10 min. Work-up as described for **1b** gave a soln of *N*-oxide (**3d**); TLC (15:1 $CHCl_3$–MeOH): R_f 0.17 (**3c**, R_f 0.77). Ac_2O (0.15 mL, 1.6 mmol) and dry pyridine (0.26 mL, 3.21 mmol) were added and the mixture was kept at rt overnight. Work-up as described for **1c** and **1d** gave a crude mixture of **3e** and **3f**, which were separated by chromatography (4:1 toluene–EtOAc, then 4:1 toluene–acetone). Concn of the fraction having R_f 0.67 (4:1 toluene–acetone) gave **3e** (466 mg, 64%) as a colorless solid; $[\alpha]_D^{22}$ −47° (*c* 1, $CHCl_3$). Anal. Calcd for $C_{57}H_{98}O_{19}Si_2$: C, 59.87; H, 8.64. Found: C, 59.95; H, 8.57.

Concn of the fraction having R_f 0.37 (4:1 toluene–acetone) gave **3f** (466 mg, 64%) as a colorless solid; $[\alpha]_D^{23}$ −44° (*c* 1, $CHCl_3$). Anal. Calcd for $C_{60}H_{105}NO_{19}Si_2$: C, 60.02; H, 8.81; N, 1.17. Found: C, 59.75; H, 8.74; N, 1.04.

2′,4′-Di-O-acetyl-3,4″-bis(O-tert-butyldimethylsilyl)-3′-de(dimethylamino)-3′-hydroxydesmycosin 9,20-bis(ethylene acetal) (**3g**).—To a cooled (−20 °C) soln of **3e** (450 mg, 0.394 mmol) in dry toluene (9 mL), 0.4 M $Zn(BH_4)_2$ in THF (1.5 mL, 0.6 mmol) was added and the mixture was kept at −20 °C for 20 min. The residue obtained after work-up as described for **1e** was purified by chromatography (7:1 toluene–acetone) to give **3g** (334 mg, 74%) as a colorless solid; $[\alpha]_D^{22}$ −58° (*c* 1, $CHCl_3$); TLC (7:1 toluene–acetone): R_f 0.22 (**3e**, R_f 0.52). Anal. Calcd for $C_{57}H_{100}O_{19}Si_2$: C, 59.76; H, 8.80. Found: C, 59.84; H, 8.78.

2′,4′-Di-O-acetyl-3,4″-bis(O-tert-butyldimethylsilyl)-3′-de(dimethylamino)-3′-methoxydesmycosin 9,20-bis(ethylene acetal) (**3h**).—To a soln of **3g** (238 mg, 0.21 mmol) in dry CH_2Cl_2 (5 mL) were added DTBP (1.85 mL, 8.25 mmol) and MeOTf (0.47 mL, 4.15 mmol) and the mixture was refluxed for 48 h. After addition of MeOH (0.169 mL, 4.17 mmol), the mixture was worked up as described above for **1f**. The residue was chromatographed (9:1 toluene–acetone) to afford **3h** (106 mg, 44%) as a colorless solid; $[\alpha]_D^{23}$ −56° (*c* 1, $CHCl_3$); TLC (9:1 toluene-acetone): R_f 0.33 (**3e**, R_f 0.12). Anal. Calcd for $C_{58}H_{102}O_{19}Si_2$: C, 60.08; H, 8.71. Found: C, 60.31; H, 8.71.

*3,4''-Bis(*O*-tert-butyldimethylsilyl)-3'-de(dimethylamino)-3'-methoxydesmycosin 9,20-bis(ethylene acetal)* (**3i**).—A soln of **3h** (62.0 mg, 0.053 mmol) in 0.1 M methanolic NaOMe (1.5 mL) was heated at 50 °C for 3 h. Work-up as described above for deacetylation of **1f** gave **3i** (46.7 mg, 81%) as a colorless solid; $[\alpha]_D^{24}$ −24° (*c* 1, $CHCl_3$); TLC (5:1 toluene–acetone): R_f 0.30 (**3h**, R_f 0.58). Anal. Calcd for $C_{54}H_{98}O_{17}Si_2$: C, 60.30; H, 9.18. Found: C, 60.33; H, 9.31.

3' - De(dimethylamino) - 3' - methoxydesmycosin (**3j**).—Compound **3i** (34.0 mg, 0.032 mmol) was dissolved in MeCN (0.5 mL), 0.1 M aq HCl (0.5 mL) was added, and the mixture was heated at 50 °C for 9 h. After work-up as described above for **1g**, purification by chromatography (3:2 $CHCl_3$–acetone) gave **3j** (12.9 mg, 54%) as a colorless solid; $[\alpha]_D^{22}$ −20° (*c* 1, $CHCl_3$); TLC (3:2 $CHCl_3$–acetone): R_f 0.27 (**3i**, R_f 0.72). FAB-MS: *m/z* 759 $(M+1)^+$. Anal. Calcd for $C_{38}H_{62}O_{15}\cdot 0.5H_2O$: C, 59.43; H, 8.27. Found: C, 59.30; H, 8.00.

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