Cladinose Analogues of Sixteen-membered Macrolide Antibiotics

IV. Improved Therapeutic Effects of 4-*O*-Acyl-L-cladinose Analogues of Sixteen-membered Macrolide Antibiotics

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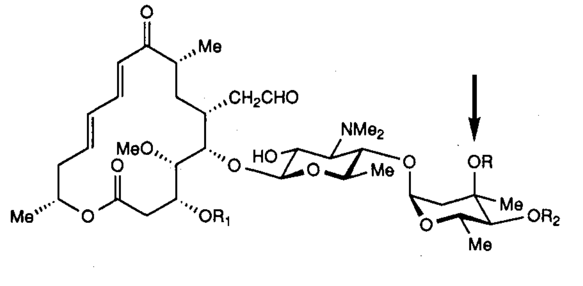
Six derivatives of sixteen-membered macrolides possessing 4-*O*-acyl-α-L-cladinose as a neutral sugar were synthesized *via* 3″-methylthiomethyl ether intermediates in reasonable yield. Introduction of a methyl group on the 3″-hydroxyl group of midecamycin A_1 was effective for enhancing its antibacterial activity. All these derivatives exhibited excellent therapeutic effects in mice, and some of them showed improved pharmacokinetics compared with the natural antibiotics (mycarose type) in mice. Facile synthesis of 9-*O*-acylated analogues are also described.

Sixteen-membered macrolide antibiotics[1)] produced by several kinds of *Streptomyces* species are used in the clinic, and the chemically modified derivatives, such as rokitamycin (RKM)[2)] and miocamycin (MOM)[3)] show good therapeutic effects. Although these chemotherapeutics belong to the leucomycin family (platenomycin skeleton), potent tylosin analogues have also been reported[4,5)].

We have been focusing our attention on leucomycin analogues because of its effectiveness and low side effects, and have reported preparations and biological activities of 4-*O*-alkyl-α-L-cladinose analogues[6~8)]. In 1977, TATSUTA *et al.*[9)] reported synthesis of a cladinose analogue of carbomycin B, compound (**1**) (Fig. 1) as a pioneer work, and showed that **1** had enhanced activity against *Mycobacterium smegmatis* in comparison with carbomycin B but comparable activity against other bacteria. Another cladinose analogue (**2**)[10)] we synthesized, however, exhibited stronger antibacterial activity against many kinds of clinically important organisms than midecamycin A_3 (Fig. 1). These results suggested that the effect of introducing a methyl group into the 3″-hydroxyl group may be different depending on the parent structure. On the other hand, pharmacokinetics of sixteen-membered derivatives possessing an sp^3 carbon at the C-9 position have been shown to be much improved *in vivo*[6,7,11)] than those of sp^2 compounds in mice. These observations prompted us to prepare and investigate 4-*O*-acyl-L-cladinose analogues with an sp^3 carbon at C-9.

In this paper, we wish to report a short synthesis of cladinose analogues (**8a** and **8b**) of midecamycin A_1 (MDM) and josamycin (JM), and facile preparation of their 9-*O*-acyl derivatives (**9a**, **9b**, **10a** and **10b**). Compound (**9a**) showed comparable antibacterial activity *in vitro* to that of MOM, but exhibited excellent therapeutic efficacy *in vivo*.

Fig. 1. Natural 9-dehydro antibiotics and their cladinose analogues.



	R_1	R_2	R
Carbomycin B	Ac	$COCH_2CH(CH_3)_2$	H
Midecamycin A_3	COEt	COEt	H
1	Ac	$COCH_2CH(CH_3)_2$	Me
2	COEt	COEt	Me

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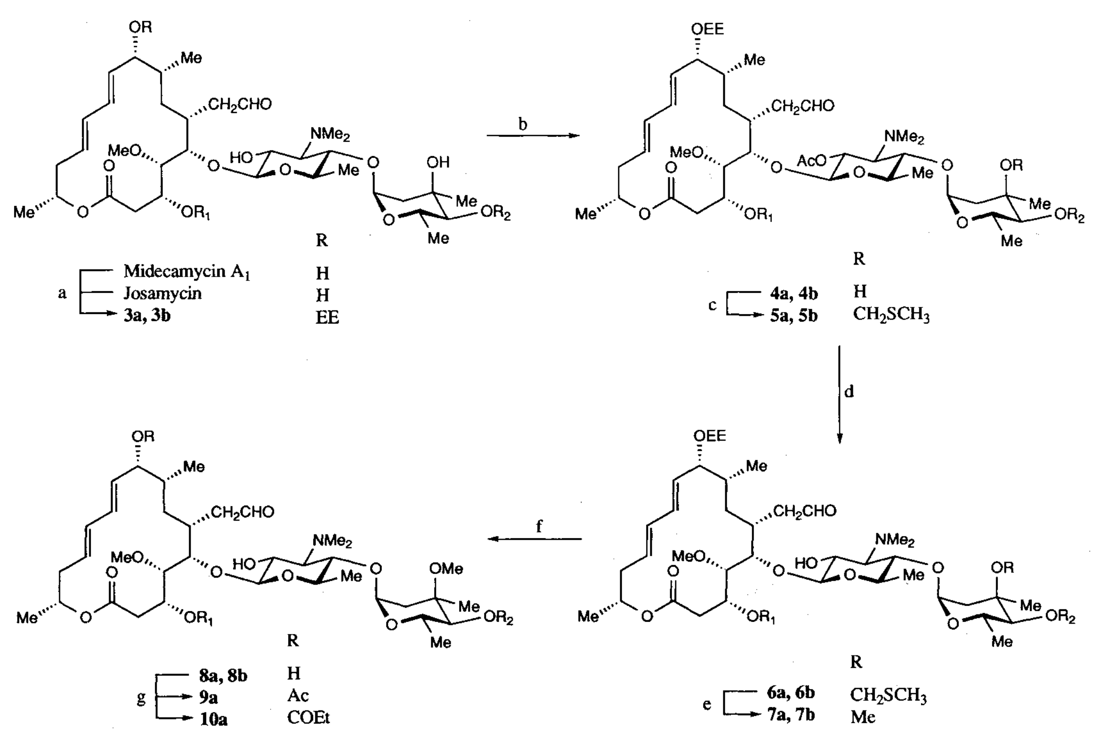
Chemistry

Since there were some difficulties at an early stage to construct a 3″-methyl ether by direct methylation in the presence of a 4″-acyl group, an unprotected aldehyde group and an unmodified lactone ring[††,8,12,13], we first selected an indirect method for introducing a methyl group into a tertiary hydroxyl group at the C-3″ position to prepare titled compounds (**8a** and **8b**) (Scheme 1). Here, we used a methylthiomethyl (MTM) ether as a key intermediate to generate a 3″-OCH_3 group. A 3″-MTM ether of MDM has been already reported[14] as a useful semisynthetic analogue of MDM.

An allylic alcohol at the C-9 position of MDM could be chemoselectively protected as its 1-ethoxyethyl (EE) ether, **3a** (Scheme 1). Acetylation of a 2′-hydroxyl group without basic catalyst gave a tertiary alcohol (**4a**) quantitatively. Methylthiomethylation with a known method[14] afforded an MTM ether (**5a**) in 70% yield, which was then deacetylated with methanol to give the key intermediate (**6a**) in high yield. An MTM ether was reported to be converted into a methyl ether *via* heterogeneous hydrogenolysis[15~17] in the end of the 1960's. This key compound (**6a**), however, possesses sensitive functional groups (double bonds and an aldehyde group) which are labile under hydrogenolysis conditions. As expected, usual hydrogenolysis of these MTM ethers easily gave perhydrogenated (10, 11, 12, 13, 18, 18-hexahydro) 3″-OCH_3 compounds. However, deactivated Raney-Nickel with the optimized method (see experimental) converted **6a** to a desired methyl ether (**7a**) in a moderate yield. In this reaction, a 3″-alcohol (**3a**) and a 3″-$OCH_2OC_2H_5$ analogue were obtained as by products even in the optimized condition. The EE group of **7a** was finally removed to prepare the titled compound, **8a**, *via* acidic conditions. Reduction of the MTM ether to a methoxy group was done at the stage before removal of the EE group, because reduction after removal of EE led to complicated results.

3″-*O*-Methyljosamycin (**8b**) was also synthesized *via* the same methodology (Scheme 1). Chemoselective acylation[18] of the allylic alcohol of **8a** afforded 9-*O*-acyl analogues, **9a** and **10a**.

Scheme 1. Synthesis of compounds **8a** and **8b**[a].



[a]Reagents and conditions: (a) $H_5C_2OCH{=}CH_2$, PPTS, CH_2Cl_2, 25°C, 16 h; (b) Ac_2O, CH_3CN, 40°C, 16 h; (c) Ac_2O, DMSO, 33°C, 64 h; (d) MeOH, 30°C, 16 h; (e) deactivated Raney-Nickel, EtOH, 25°C, 20 min.; (f) AcOH, aqueous CH_3CN, 25°C, 16 h; (g) AcCl or EtCOCl, Pyr., PhMe, 25°C, 1 h.
EE: 1-Ethoxyethyl. Midecamycin A_1 derivatives are represented as suffix "a" compounds; $R_1 = R_2 =$ COEt.
Josamycin derivatives are represented as suffix "b" compounds; $R_1 =$ Ac, $R_2 = COCH_2CH(CH_3)_2$.

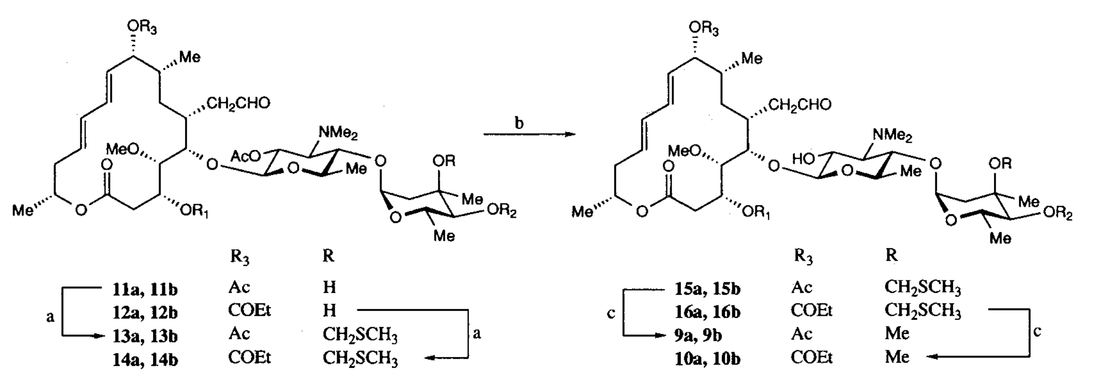
†† Some intermediates with a protected aldehyde and a modified lactone ring are available for further reactions. See ref. 12 and 13. A direct methylation at the C-3″ position could be done under stronger conditions. See ref. 8.

To prepare the above mentioned 9-*O*-acyl derivatives (**9**, **10**), a more facile synthetic method (Scheme 2) was also used. Fully protected tertiary alcohols (**11a**, **11b**, **12a** and **12b**) were methylthiomethylated followed by deacetylation at the C-2′ position to give key intermediates, **15a**, **15b**, **16a** and **16b**. Optimized hydrogenolysis of the MTM ethers consequently afforded 9-*O*-acyl-3″-*O*-methylmidecamycin A_1 (**9a** and **10a**) and 9-*O*-acyl-3″-*O*-methyljosamycin (**9b** and **10b**). Thus, this efficient short synthetic route enabled us to prepare these analogues very easily for *in vivo* studies.

Biological Evaluation

Antibacterial activities *in vitro* of the novel 4-*O*-acyl-α-L-cladinosyl derivatives (**8a**, **8b**, **9a**, **9b**, **10a** and **10b**), compared with those of natural antibiotics, MDM, JM, and semisynthetic MOM, are shown in Table 1. As judged from the MIC values, 3″-*O*-methylmidecamycin A_1, **8a**, exhibited about two-fold higher activity than that of MDM against almost all kinds of organisms compared. On the other hand, 3″-*O*-methylation of JM did not increase its *in vitro* activity (**8b** *vs.* JM). These observations suggested that it might be possible to optimize the 4″-*O*-acyl group for the highest antibacterial

Scheme 2. Facile synthesis of 9-*O*-acyl derivatives[a].



[a]Reagents and conditions: (a) Ac_2O, DMSO, 33°C, 64 h; (b) MeOH, 30°C, 16 h; (c) deactivated Raney-Nickel, EtOH, 25°C, 20 min.
Midecamycin A_1 derivatives are represented as suffix "a" compounds; $R_1 = R_2 =$ COEt.
Josamycin derivatives are represented as suffix "b" compounds; R_1 = Ac, $R_2 = COCH_2CH(CH_3)_2$.

Table 1. Antibacterial activities of 4-*O*-acyl-L-cladinose analogues and reference chemotherapeutics (MIC, μg/ml).

Test Organisms	**8a**	**8b**	**9a**	**9b**	**10a**	**10b**	MDM	JM	MOM
Staphylococcus aureus 209P JC-1	0.20	0.10	0.20	0.20	0.39	0.39	0.39	0.20	0.20
S. aureus M133	0.39	0.78	0.78	0.78	1.56	1.56	0.78	0.78	0.78
S. aureus M126	>100	>100	>100	>100	>100	>100	>100	>100	>100
S. aureus MS15026	>100	>100	>100	>100	>100	>100	>100	>100	>100
S. aureus MS15027	0.39	0.78	0.78	0.78	1.56	1.56	0.78	0.78	0.78
S. epidermidis ATCC14990	0.78	0.78	0.78	0.78	1.56	1.56	0.78	0.78	0.78
Micrococcus luteus ATCC9341	0.05	0.05	0.10	0.10	0.10	0.20	0.05	0.05	0.10
Enterococcus faecalis W-73	1.56	3.13	1.56	1.56	3.13	3.13	3.13	3.13	1.56
Streptococcus pneumoniae IP692	0.10	0.20	0.10	0.20	0.20	0.78	0.39	0.10	0.20
S. pneumoniae Type I	0.10	0.20	0.10	0.20	0.39	0.78	0.39	0.10	0.20
S. pyogenes Cook	0.10	0.10	0.20	0.20	0.20	0.20	0.20	0.10	0.20
Escherichia coli NIHJ JC-2	>100	>100	>100	>100	>100	>100	>100	>100	>100
Klebsiella pneumoniae PCI602	>100	>100	>100	>100	>100	>100	>100	>100	>100
Moraxella catarrhalis W-0500	0.78	0.78	1.56	1.56	1.56	1.56	3.13	0.78	1.56
M. catarrhalis W-0506	0.78	0.78	1.56	1.56	6.25	6.25	3.13	0.78	1.56
Haemophilus influenzae 9334	1.56	1.56	3.13	6.25	6.25	12.5	6.25	1.56	6.25
H. influenzae Type b	12.5	12.5	25	50	50	50	25	12.5	25

Table 2. Protective effects of 4-*O*-acyl-α-L-cladinose analogues and MOM on systemic infections in mice.

Organisms	Inoculum size[a] (CFU/mouse)	Compound[b]	ED_{50}[c] (mg/kg)	MIC (μg/ml)
Staphylococcus aureus Smith I	7.8×10^6	**8a**	1.7×10^2	0.78
		8b	1.1×10^2	0.78
		9a	1.0×10^2	0.78
		9b	0.9×10^2	1.56
		10a	1.1×10^2	1.56
		10b	1.3×10^2	1.56
		MOM	4.6×10^2	1.56
Streptococcus pneumoniae DP-I Type I	1.6×10^2	**8a**	4.9×10^1	0.20
		8b	4.0×10^1	0.39
		9a	1.7×10^1	0.20
		9b	1.8×10^1	0.39
		10a	1.5×10^1	0.20
		10b	4.5×10^1	0.78
		MOM	6.7×10^1	0.20

[a] Administered intraperitoneally with 2.5% gastric mucin.
[b] Single oral dosing one hour after infection.
[c] ED_{50}, 50% effective dose.

activity. 9-*O*-Acetylation of **8a** affected only slightly its *in vitro* activity, and compound (**9a**) was comparable to MOM in activity.

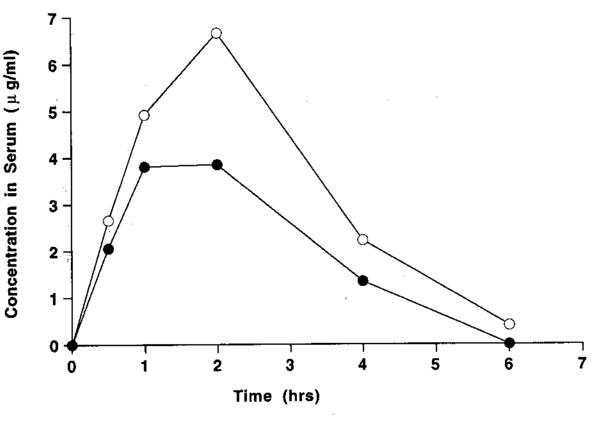
Next, we examined the duration of activity of **8a** and **8b** which were structurally fundamental compounds. Incubation with rat plasma *in vitro*††† revealed that the half-life ($T_{1/2}$) of 4-*O*-acyl-L-cladinose analogues (**8a** and **8b**) was about two-fold times longer than that of 4-*O*-acyl-L-mycarose counterparts (MDM and JM), respectively (data not shown). It was found furthermore that a deacylated metabolite (4-OH-cladinoside) of **8a** and **8b** was more potent than that (4-OH-mycaroside)[19] of the natural antibiotic (data will be published in a separate paper). These promising characters of the novel 4-*O*-acyl-L-cladinose analogues prompted us to investigate their *in vivo* activity.

The protective effects of these 3″-*O*-methyl sixteen-membered macrolide antibiotics on systemic infections in mice are shown in Table 2. As judged from the ED_{50} values, the *in vivo* activities against *Staphylococcus aureus* Smith I of these new six analogues, especially **9a** and **9b** were four times or more potent than that of MOM. Compounds **9a**, **9b** and **10a** also were about four times more potent than MOM in protective effects against *Streptococcus pneumoniae* DP-I type I.

Pharmacokinetics of **9a** as a representative analogue of these new antibiotics were studied preliminarily to clarify the mechanisms of their excellent *in vivo* effects.

Fig. 2. Time course of serum concentrations of **9a** and MOM after oral dosing (200 mg/kg) in mice (n=4).

○ **9a**, ● MOM.



Serum concentrations of **9a** and MOM after oral administration are shown in Fig. 2. Compound (**9a**) exhibited a higher serum level than that of MOM. Furthermore, urinary excretion of **9a** was greatly improved compared with MOM and MDM as shown in Fig. 3. Especially noted was large accumulation of **9a** 4 hours after dosing, indicating the delayed excretion of this compound as compared with the references. This analogue (**9a**) was distributed in lung more than in serum in mice (Fig. 4), similar to MOM in rat[19]. Metabolic studies in mice after oral administration of compound

††† Metabolic studies using rat plasma were done as described in our previous paper. See ref. 6 or 8.

Fig. 3. Time course of urinary recovery of **9a**, MOM and MDM after oral dosing (200 mg/kg) in mice (n = 6).

■ 4～24 hours, ▨ 2～4 hours, ▧ 0～2 hours.

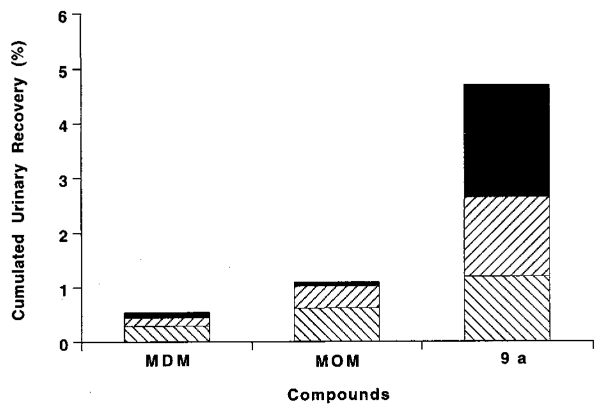
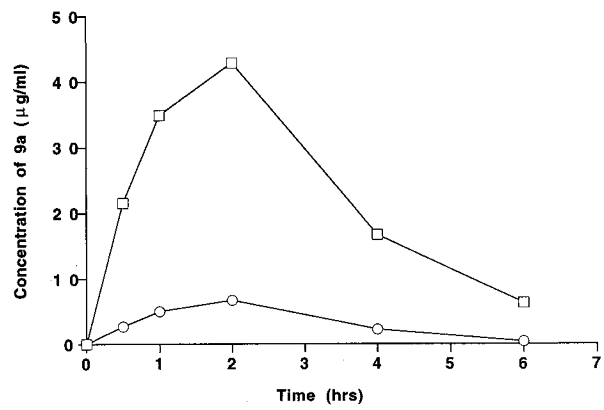


Fig. 4. Time course of distribution of **9a** in serum and lung after oral dosing (200 mg/kg) in mice (n = 4).

○ Serum, □ lung.



(**9a**), revealed that one of the main metabolites was 3″-*O*-methyl-4″-de-*O*-propionylmidecamycin A_1 which was more potent *in vitro* than 4″-de-*O*-propionylmidecamycin A_1. The excellent *in vivo* effects of **9a** and its related analogues, compared with those of the C-3″ hydroxyl-counterparts, are related most probably to with higher serum levels, increased urinary excretion and more potent activity of a metabolite.

In conclusion, a series of sixteen-membered macrolides possessing a 4-*O*-acyl-α-L-cladinose moiety were synthesized *via* 3″-methylthiomethyl intermediates. They showed antibacterial activity *in vitro* comparable to or two～four-fold more potent than the counterparts possessing a 4-*O*-acyl-α-L-mycarose moiety, but more potent *in vivo* activity, supported by better pharmacokinetic profile and a potent metabolite.

Experimental

General Methods

MP's were determined with a Yanagimoto micro melting point apparatus and were uncorrected. Optical rotations were measured on a Perkin-Elmer 241 polarimeter. Mass spectra were obtained on a Hitachi M-80A or M-80B mass spectrometer for EI-MS or FD-, SI-MS, respectively. ^{1}H NMR spectra were measured with a Jeol JNM-GSX 400 NMR spectrometer for 400 MHz in $CDCl_3$ using TMS as an internal standard. Silica gel chromatography and preparative TLC were performed on Merck Kieselgel 60 and Merck TLC $60F_{254}$, respectively. In general, the organic layer was dried with anhydrous Na_2SO_4, evaporation and concentration were carried out under reduced pressure below 30°C, unless otherwise noted.

Antibacterial Activity *In Vitro*

Minimum inhibitory concentrations (MICs) were determined by the agar dilution method. Test strains were subjected to seed culture using Sensitivity test broth (STB, Nissui Pharmaceutical) except the strains belonging to the genus *Streptococcus*, *Moraxella* and *Haemophilus* which were cultured on blood agar plate. A 5 μl portion of cell suspension of the test strains having about 10^6 CFU/ml was inoculated into Sensitivity disk agar (SDA, Nissui Pharmaceutical) supplemented with 5% horse blood in cases of *Streptococcus*, *Moraxella* and *Haemophilus* sp. After incubation at 37°C for 20 hours, MICs were determined.

Determination of *In Vivo* Activity

Six male ICR mice weighing 19 to 21 g were used for each dose. Bacterial cells cultured overnight on heart infusion agar (Nissui Seiyaku) at 37°C were suspended in saline, and mixed with an equal volume of 5% gastric mucin (Difco). A 0.5 ml volume of bacterial suspension was inoculated intraperitoneally. Under these conditions, untreated mice died within 48 hours. The test compounds were suspended in 0.2% CMC and given as a single oral dose to the mice one hour after infection. The numbers of survivors were recorded on day 3 after infection. The 50% effective doses were calculated by the probit method[20].

Pharmacokinetics Tests in Mice

A test compound was suspended with a 0.2% aqueous solution of CMC to give a concentration of 4.0 mg/ml, and a 1.0 ml portion was orally administered to 4 weeks old male Jcl : ICR mice. Blood was collected from the axillary artery of the mice 0.5, 1, 2 ,4 and 6 hours after the administration of the test compound (n = 4). The collected blood was allowed to stand at 0°C for 2 hours and centrifuged at 3000 rpm for 20 minutes to obtain serum. To the serum was added an equivalent volume of 50% CH_3CN-0.05 M phosphate buffer (pH 7.0). The concentration of the test compound in the resulting serum sample was measured by a bioassay method using *M. luteus* ATCC9341 as a test organism.

Separately, 200 mg/kg of a test compound was orally administered to six mice in the same manner as described above. The mice were put in a metabolic cage MM type (Sugiyamagen Co., Tokyo, Japan) and urine was collected 2, 4 and 24 hours after the administration. The collected urine was filtered through a Millipore filter having a pore size of 0.45 mm and was mixed with an equivalent volume of 50% CH_3CN-0.05 M phosphate buffer (pH 6.5) to serve as a urine sample. The bioassay was carried out by *M. luteus* to as described above, and the recovery in the urine was calculated.

The concentration of the test compound in the lung was determined by the published method[21].

9-*O*-(1-Ethoxyethyl)midecamycin A_1 (**3a**)

To a solution of MDM (20.0 g, 24.6 mmol) and ethyl vinyl ether (22 ml, 0.23 mol) in dry CH_2Cl_2 (600 ml) was added pyridinium *p*-toluenesulfonate (PPTS) (9.40 g, 37.4 mmol). The solution was kept at room temperature for 16 hours, and then poured into saturated aqueous $NaHCO_3$ (2.0 liters). The mixture was extracted with $CHCl_3$ (1.8 liters), and the organic layer was successively washed with 5% aqueous $NaHCO_3$ (2.0 liters) and brine (2.0 liters). After drying the organic layer, evaporation gave a residue which was purified by silica gel column chromatography (1.0 kg, $CHCl_3$ - MeOH, 50 : 1) to afford **3a** (20.0 g, 92%) as a colorless solid: MP 100~103°C; $[\alpha]_D^{21}$ −61° (*c* 1.0, MeOH); EI-MS *m/z* 885 (M^+); ^1H NMR δ 0.99 and 1.00 (3H, 2×d, 19-H), 1.12 (3H, s, 3″-CH_3), 1.13 (3H, d, 6″-H), 1.18 (3H, t, 4″-$OCOCH_2CH_3$), 1.44 (1H, br dt, 7-H), 1.85 (1H, dd, 2″-Hax), 1.90 (1H, m, 8-H), 2.01 (1H, d, 2″-Heq), 2.15 (1H, br dt, 14-H), 2.28 (1H, br d, 2-H), 2.44 and 2.46 (2H, 2×q, 4″-$OCOCH_2CH_3$), 2.51 (6H, s, 3′-$N(CH_3)_2$), 2.63 (1H, br dq, 3-$OCOCH_2CH_3$), 2.73 and 2.74 (1H, 2×dd, 2-H), 2.82 and 2.83 (1H, 2×br dd, 17-H), 3.24 (1H, br d, 4-H), 3.44 and 3.63 (2H, 2×dq, 9-$OCH(OCH_2CH_3)CH_3$), 3.53 (3H, s, 4-OCH_3), 3.78 and 3.92 (1H, 2×dd, 9-H), 3.87 (1H, br d, 5-H), 4.41 (1H, d, 1′-H), 4.46 (1H, dq, 5″-H), 4.62 (1H, d, 4″-H), 4.64 and 4.66 (1H, 2×q, 9-$OCH(OCH_2CH_3)CH_3$), 5.02 (1H, ddq, 15-H), 5.07 (1H, d, 1″-H), 5.13 (1H, br d, 3-H), 5.47 and 5.56 (1H, 2×dd, 10-H), 5.78 and 5.82 (1H, 2×br ddd, 13-H), 6.09 (1H, br dd, 12-H), 6.57 and 6.61 (1H, 2×dd, 11-H), 9.64 and 9.65 (1H, 2×br s, 18-H).

9-*O*-(1-Ethoxyethyl)josamycin (**3b**)

Reaction of JM with ethyl vinyl ether gave **3b** as a colorless solid in 90% yield by a similar procedure to **3a**.

3b: MP 105~108°C; $[\alpha]_D^{16}$ −68° (*c* 1.0, MeOH); EI-MS *m/z* 899 (M^+); ^1H NMR δ 0.98 and 0.99 (3H, 2×d, 19-H), 0.98 (6H, d, 4″-$OCOCH_2CH(CH_3)_2$), 1.12 (3H, s, 3″-CH_3), 1.14 (3H, d, 6″-H), 1.14 (3H, t, 9-$OCH(OCH_2CH_3)CH_3$), 1.19 (3H, d, 6′-H), 1.23 and 1.25 (3H, 2×d, 9-$OCH(OCH_2CH_3)CH_3$), 1.27 (3H, d, 16-H), 1.45 (1H, br dt, 7-H), 1.85 (1H, dd, 2″-Hax), 1.90 (1H, m, 8-H), 2.02 (1H, d, 2″-Heq), 2.52 (6H, s, 3′-$N(CH_3)_2$), 2.73 and 2.75 (1H, 2×dd, 2-H), 2.84 and 2.85 (1H, br dd, 17-H), 3.24 (1H, br d, 4-H), 3.28 (1H, t, 4′-H), 3.35, 3.42 and 3.63 (2H, 3×dq, 9-$OCH(OCH_2CH_3)CH_3$), 3.54 (3H, s, 4-OCH_3), 3.75 and 3.89 (1H, 2×dd, 9-H), 3.90 (1H, br d, 5-H), 4.43 (1H, d, 1′-H), 4.46 (1H, dq, 5″-H), 4.63 (1H, d, 4″-H), 4.63 and 4.64 (1H, 2×q, 9-$OCH(OCH_2CH_3)CH_3$), 5.04 (1H, ddq, 15-H), 5.07 (1H, d, 1″-H), 5.12 (1H, br d, 3-H), 5.46 and 5.55 (1H, 2×dd, 10-H), 5.76 and 5.81 (1H, 2×ddd, 13-H), 6.08 (1H, br dd, 12-H), 6.56 and 6.58 (1H, 2×dd, 11-H), 9.64 and 9.65 (1H, 2×s, 18-H).

2′-*O*-Acetyl-9-*O*-(1-ethoxyethyl)midecamycin A_1 (**4a**)

To a solution of **3a** (12.5 g, 14.1 mmol) in dry CH_3CN (370 ml) was added acetic anhydride (2.7 ml, 29 mmol). The solution was kept at 40°C for 16 hours and cooled down to room temperature. After 1 N NH_4OH (42 ml, 42 mmol) was added, the mixture was allowed to stand at room temperature for 10 minutes. It was concentrated to give a residue which was extracted with $CHCl_3$ (1.0 liter). The organic layer was washed with saturated aqueous $NaHCO_3$ (1.0 liter) and brine (1.2 liters). After drying the organic layer, evaporation gave **4a** (13.0 g, 99%) as a colorless solid: MP 104~107°C; $[\alpha]_D^{21}$ −64° (*c* 1.0, $CHCl_3$); SI-MS *m/z* 928 (MH^+); ^1H NMR δ 0.98 and 0.99 (3H, 2×d, 19-H), 1.12 (3H, s, 3″-CH_3), 1.18 (3H, t, 4″-$OCOCH_2CH_3$), 1.42 (1H, br dt, 7-H), 1.84 (1H, dd, 2″-Hax), 2.00 (1H, d, 2″-Heq), 2.02 (3H, s, 2′-$OCOCH_3$), 2.15 (1H, br dt, 14-H), 2.24 (1H, br d, 2-H), 2.26 and 2.81 (1H, 2×br dd, 17H), 2.41 (6H, s, 3′-$N(CH_3)_2$), 2.43 and 2.44 (2H, 2×q, 4″-$OCOCH_2CH_3$), 2.65 (1H, dq, 3-$OCOCH_2CH_3$), 2.70 (1H, dd, 2-H), 2.81 (1H, br dd, 17-H), 3.17 (1H, br d, 4-H), 3.35, 3.43, 3.49 and 3.62 (2H, 4×dq, 9-$OCH(OCH_2CH_3)CH_3$), 3.47 (3H, s, 4-OCH_3), 3.75 and 3.88 (1H, 2×dd, 9-H), 3.89 (1H, br d, 5-H), 4.37 (1H, dq, 5″-H), 4.61 (1H, d, 1′-H), 4.62 (1H, d, 4″-H), 4.64 and 4.65 (1H, 2×q, 9-$OCH(OCH_2CH_3)CH_3$), 4.98 (1H, dd, 2′-H), 5.06 (1H, d, 1″-H), 5.11 (1H, br d, 3-H), 5.45 and 5.54 (1H, 2×dd, 10-H), 5.79 and 5.83 (1H, 2×ddd, 13-H), 6.05 (1H, br dd, 12-H), 6.57 and 6.61 (1H, 2×dd, 11-H), 9.62 and 9.63 (1H, 2×br s, 18-H).

2′-*O*-Acetyl-9-*O*-(1-ethoxyethyl)josamycin (**4b**)

Reaction of **3b** with acetic anhydride gave **4b** as a colorless solid in 96% yield by a similar procedure to **4a**.

4b: MP 110~113°C; $[\alpha]_D^{19}$ −70° (*c* 1.0, $CHCl_3$); FD-MS *m/z* 942 (MH^+); ^1H NMR δ 0.86 (1H, br ddd, 7-H), 0.98 (6H, d, 4″-$OCOCH_2CH(CH_3)_2$), 1.12 (3H, s, 3″-CH_3), 1.13 (3H, d, 6″-H), 1.14 (3H, t, 9-$OCH(OCH_2CH_3)CH_3$), 1.18 (3H, d, 6′-H), 1.22 and 1.24 (3H, 2×d, 9-$OCH(OCH_2CH_3)CH_3$), 1.26 (3H, d, 16-H), 1.43 (1H, br dt, 7-H), 1.84 (1H, dd, 2″-Hax), 2.01 (1H, d, 2″-Heq), 2.02 (3H, s, 2′-$OCOCH_3$), 2.12 (1H, dt, 14-H), 2.25 (1H, br d, 2-H), 2.29 (3H, s, 3-$OCOCH_3$), 2.41 (6H, s, 3′-$N(CH_3)_2$), 2.45 (1H, br dt, 14-H), 2.69

(1H, t, 3′-H), 2.72 (1H, dd, 2-H), 2.83 (1H, br dd, 17-H), 3.18 (1H, br d, 4-H), 3.34, 3.41 and 3.63 (2H, 3 × dq, 9-OCH(OC*H*$_2$CH$_3$)CH$_3$), 3.49 (3H, s, 4-OCH$_3$), 3.72 and 3.86 (1H, 2 × dd, 9-H), 3.91 (1H, br d, 5-H), 4.38 (1H, dq, 5″-H), 4.99 (1H, dd, 2′-H), 5.01 (1H, ddq, 15-H), 5.06 (1H, d, 1″-H), 5.10 (1H, br d, 3-H), 5.46 and 5.54 (1H, 2 × dd, 10-H), 5.76 and 5.81 (1H, 2 × ddd, 13-H), 6.05 (1H, br dd, 12-H), 6.55 and 6.58 (1H, 2 × dd, 11-H), 9.63 and 9.64 (1H, 2 × s, 18-H).

2′-*O*-Acetyl-9-*O*-(1-ethoxyethyl)-3″-*O*-(methylthiomethyl)midecamycin A$_1$ (**5a**)

A solution of **4a** (3.05 g, 3.29 mmol) in dry DMSO (91 ml) and acetic anhydride (9.1 ml, 96 mmol) was kept at 33°C for 64 hours, then poured into toluene (600 ml). The organic layer was washed with H_2O (600 ml) three times, and dried. Evaporation gave a residue which was purified by silica gel column chromatography (300 g, hexane - EtOAc, 1 : 1) to afford **5a** (2.27 g, 70%) as a colorless solid and recovered **4a** (549 mg, 18%). This diastereoisomeric mixture **5a** was separated by preparative TLC (hexane - EtOAc, 1 : 1) to obtain a less polar isomer (**5aH**) (1.34 g) and a more polar isomer (**5aL**) (0.93 g).

5aH: MP 94 ~ 96°C; $[\alpha]_D^{19}$ −71° (*c* 1.0, CHCl$_3$); FD-MS *m/z* 988 (MH$^+$); ^{1}H NMR δ 0.85 (1H, br dt, 7-H), 0.98 (3H, d, 19-H), 1.05 (3H, d, 6″-H), 1.14 (3H, t, 9-OCH(OCH$_2$C*H*$_3$)CH$_3$), 1.14 (3H, d, 6′-H), 1.17 (3H, s, 3″-CH$_3$), 1.18 (3H, t, 4″-OCOCH$_2$C*H*$_3$), 1.21 (3H, t, 3-OCOCH$_2$C*H*$_3$), 1.22 (3H, d, 9-OCH(OCH$_2$CH$_3$)C*H*$_3$), 1.26 (3H, d, 16-H), 1.41 (1H, br dt, 7-H), 1.68 (1H, dd, 2″-Hax), 1.87 (1H, m, 8-H), 2.01 (3H, s, 2′-OCOCH$_3$), 2.15 (1H, dt, 14-H), 2.20 (3H, s, 3″-OCH$_2$SC*H*$_3$), 2.24 (1H, br d, 2-H), 2.25 (1H, d, 2″-Heq), 2.42 (2H, q, 4″-OCOC*H*$_2$CH$_3$), 2.43 (6H, s, 3′-N(CH$_3$)$_2$), 2.50 and 2.65 (2H, 2 × dq, 3-OCOC*H*$_2$CH$_3$), 2.68 (1H, t, 3′-H), 2.72 (1H, dd, 2-H), 2.83 (1H, br dd, 17-H), 3.16 (1H, t, 4′-H), 3.17 (1H, br d, 4-H), 3.26 (1H, dq, 5′-H), 3.42 and 3.50 (2H, 2 × dq, 9-OCH(OC*H*$_2$CH$_3$)CH$_3$), 3.49 (3H, s, 4-OCH$_3$), 3.87 (1H, br d, 5-H), 3.88 (1H, dd, 9-H), 4.51 (1H, d, 3″-OC*H*$_2$SCH$_3$), 4.56 (1H, dq, 5″-H), 4.60 (1H, d, 1′-H), 4.64, 4.65 (2H, 2 × d, 4″-H, 3″-OC*H*$_2$SCH$_3$), 4.81 (1H, d, 1″-H), 4.92 (1H, dd, 2′-H), 4.99 (1H, ddq, 15-H), 5.11 (1H, br d, 3-H), 5.45 (1H, dd, 10-H), 5.82 (1H, ddd, 13-H), 6.06 (1H, br dd, 12-H), 6.61 (1H, dd, 11-H), 9.62 (1H, br s, 18-H).

5aL: MP 90 ~ 94°C; $[\alpha]_D^{19}$ −87° (*c* 1.0, CHCl$_3$); FD-MS *m/z* 988 (MH$^+$); ^{1}H NMR δ 0.87 (1H, br dt, 7-H), 0.98 (3H, d, 19-H), 1.05 (3H, d, 6″-H), 1.14 (3H, t, 9-OCH(OCH$_2$C*H*$_3$)CH$_3$), 1.14 (3H, d, 6′-H), 1.17 (3H, s, 3″-CH$_3$), 1.18 (3H, t, 4″-OCOCH$_2$C*H*$_3$), 1.23 (3H, t, 3-OCOCH$_2$C*H*$_3$), 1.25 (3H, d, 9-OCH(OCH$_2$CH$_3$)-C*H*$_3$), 1.26 (3H, d, 16-H), 1.43 (1H, br dt, 7-H), 1.69 (1H, dd, 2″-Hax), 1.86 (1H, m, 8-H), 2.01 (3H, s, 2′-OCOCH$_3$), 2.15 (1H, dt, 14-H), 2.20 (3H, s, 3″-OCH$_2$SC*H*$_3$), 2.25 (1H, br d, 2-H), 2.26 (1H, d, 2″-Heq), 2.42 (2H, q, 4″-OCOC*H*$_2$CH$_3$), 2.43 (6H, s, 3′-N(CH$_3$)$_2$), 2.52 and 2.65 (2H, 2 × dq, 3-OCOC*H*$_2$CH$_3$), 2.68 (1H, t, 3′-H), 2.72 (1H, dd, 2-H), 2.84 (1H, br dd, 17-H), 3.16 (1H, t, 4′-H), 3.18 (1H, br d, 4-H), 3.26 (1H, dq, 5′-H), 3.35 and 3.63 (2H, 2 × dq, 9-OCH(OC*H*$_2$CH$_3$)CH$_3$), 3.49 (3H, s, 4-OCH$_3$), 3.74 (1H, dd, 9-H), 3.87 (1H, br d, 5-H), 4.51 (1H, d, 3″-OC*H*$_2$SCH$_3$), 4.57 (1H, dq, 5″-H), 4.60 (1H, d, 1′-H), 4.64, 4.65 (2H, 2 × d, 4″-H, 3″-OC*H*$_2$SCH$_3$), 4.65 (1H, q, 9-OC*H*(OCH$_2$CH$_3$)CH$_3$), 4.81 (1H, d, 1″-H), 4.92 (1H, dd, 2′-H), 5.00 (1H, ddq, 15-H), 5.11 (1H, br d, 3-H), 5.54 (1H, dd, 10-H), 5.80 (1H, ddd, 13-H), 6.06 (1H, br dd, 12-H), 6.58 (1H, dd, 11-H), 9.63 (1H, br s, 18-H).

2′-*O*-Acetyl-9-*O*-(1-ethoxyethyl)-3″-*O*-(methylthiomethyl)josamycin (**5b**)

Reaction of **4b** with DMSO gave **5b** as a colorless solid in 66% yield by a similar procedure to **5a**.

5b: MP 102 ~ 105°C; $[\alpha]_D^{16}$ −87° (*c* 1.0, CHCl$_3$); FD-MS *m/z* 1002 (MH$^+$); ^{1}H NMR δ 0.85 (1H, br ddd, 7-H), 0.98 (6H, d, 4″-OCOCH$_2$CH(C*H*$_3$)$_2$), 0.99 (3H, d, 19-H), 1.05 (3H, d, 6″-H), 1.14 (3H, br t, 9-OCH(OCH$_2$C*H*$_3$)CH$_3$), 1.14 (3H, d, 6′-H), 1.18 (3H, s, 3″-CH$_3$), 1.22 and 1.24 (3H, 2 × d, 9-OCH(OCH$_2$-CH$_3$)C*H*$_3$), 1.26 (3H, d, 16-H), 1.43 (1H, br dt, 7-H), 1.68 (1H, dd, 2″-Hax), 1.87 (1H, m, 8-H), 2.01 (3H, s, 2′-OCOCH$_3$), 2.15 (1H, dt, 14-H), 2.20 (3H, s, 3″-OCH$_2$SC*H*$_3$), 2.25 (1H, br d, 2-H), 2.42 (6H, s, 3′-N(CH$_3$)$_2$), 2.45 (1H, br dt, 14-H), 2.68 (1H, t, 3′-H), 2.72 and 2.73 (1H, 2 × dd, 2-H), 2.85 (1H, br dd, 17-H), 3.16 (1H, t, 4′-H), 3.18 (1H, br d, 4-H), 3.27 (1H, dq, 5′-H), 3.35, 3.41 and 3.63 (2H, 3 × dq, 9-OCH(OC*H*$_2$-CH$_3$)CH$_3$), 3.50 and 3.51 (3H, 2 × s, 4-OCH$_3$), 3.73 and 3.86 (1H, 2 × dd, 9-H), 3.89 (1H, br d, 5-H), 4.50 (1H, d, 3″-OC*H*$_2$SCH$_3$), 4.57 (1H, dq, 5″-H), 4.64, 4.65 (2H, 2 × d, 4″-H, 3″-OC*H*$_2$SCH$_3$), 4.81 (1H, d, 1″-H), 4.93 (1H, dd, 2′-H), 5.01 (1H, ddq, 15-H), 5.10 (1H, br d, 3-H), 5.10 (1H, d, 1′-H), 5.45 and 5.54 (1H, 2 × dd, 10-H), 5.76 and 5.82 (1H, 2 × ddd, 13-H), 6.05 (1H, br dd, 12-H), 6.54 and 6.58 (1H, 2 × dd, 11-H), 9.63 and 9.64 (1H, s, 18-H).

9-*O*-(1-Ethoxyethyl)-3″-*O*-(methylthiomethyl)midecamycin (**6a**)

A solution of **5a** (500 mg, 0.506 mmol) in MeOH (15 ml) was allowed to stand at 30°C for 16 hours. Evaporation gave a residue which was purified by preparative TLC (hexane - EtOAc, 1 : 1) to afford **6a** (440 mg, 92%) as a colorless solid.

An isomer of **6a** derived from **5aH**: MP 95 ~ 98°C; $[\alpha]_D^{19}$ −55° (*c* 1.0, MeOH); SI-MS *m/z* 946 (MH$^+$); ^{1}H NMR δ 0.93 (1H, br ddd, 7-H), 0.98 (3H, d, 19-H), 1.08 (3H, d, 6″-H), 1.53 (1H, br dt, 7-H), 1.75 (1H, dd, 2″-Hax), 1.89 (1H, m, 8-H), 2.19 (3H, s, 3″-OCH$_2$SC*H*$_3$), 2.26 (1H, br d, 2-H), 2.28 (1H, d, 2″-Heq), 2.42 (2H, q, 4″-OCOC*H*$_2$CH$_3$), 2.51 and 2.64 (2H, 2 × dq, 3-OCOC*H*$_2$CH$_3$), 2.58 (6H, s, 3′-N(CH$_3$)$_2$), 2.75 (1H, dd, 2-H), 2.85 (1H, br dd, 17-H), 3.26 (1H, br d, 4-H), 3.28 (1H, dq, 5′-H), 3.41 (1H, t, 4′-H), 3.43 and 3.50 (2H, 2 × dq, 9-OCH(OC*H*$_2$CH$_3$)CH$_3$), 3.57 (3H, s, 4-OCH$_3$), 3.88 (1H, br d, 5-H), 3.90 (1H, dd, 9-H), 4.51 (1H, d, 1'-H), 4.52 (1H, d, 3''-OCH_2SCH_3), 4.56 (1H, dq, 5''-H), 4.64 (1H, q, 9-OC*H*(OCH_2CH_3)CH_3), 4.65, 4.66 (2H, 2×d, 4''-H, 3''-OCH_2SCH_3), 4.92 (1H, d, 1''-H), 5.01 (1H, ddq, 15-H), 5.13 (1H, br d, 3-H), 5.46 (1H, dd, 10-H), 5.83 (1H, ddd, 13-H), 6.10 (1H, br dd, 12-H), 6.62 (1H, dd, 11-H), 9.63 (1H, br s, 18-H).

An isomer of **6a** derived from **5aL**: MP 91~93°C; $[\alpha]_D^{19}$ −73° (*c* 1.0, MeOH); SI-MS *m/z* 946 (MH^+); ^{1}H NMR δ 0.94 (1H, br ddd, 7-H), 1.08 (3H, d, 6''-H), 1.53 (1H, br dt, 7-H), 1.75 (1H, dd, 2''-Hax), 1.88 (1H, m, 8-H), 2.19 (3H, s, 3''-OCH_2SCH_3), 2.25 (1H, br dd, 17-H), 2.26 (1H, br d, 2-H), 2.28 (1H, d, 2''-Heq), 2.42 (2H, q, 4''-OCOCH_2CH_3), 2.52 and 2.65 (2H, 2×dq, 3-OCOCH_2CH_3), 2.59 (6H, s, 3'-N$(CH_3)_2$), 2.75 (1H, dd, 2-H), 2.85 (1H, br dd, 17-H), 3.26 (1H, br d, 4-H), 3.28 (1H, dq, 5'-H), 3.36 and 3.63 (2H, 2×dq, 9-OCH(OCH_2CH_3)CH_3), 3.41 (1H, t, 4'-H), 3.57 (3H, s, 4-OCH_3), 3.76 (1H, dd, 9-H), 3.88 (1H, br d, 5-H), 4.51 (1H, d, 1'-H), 4.52 (1H, d, 3''-OCH_2SCH_3), 4.56 (1H, dq, 5''-H), 4.65, 4.66 (2H, 2×d, 4''-H, 3''-OCH_2SCH_3), 4.92 (1H, d, 1''-H), 5.02 (1H, ddq, 15-H), 5.13 (1H, br d, 3-H), 5.55 (1H, dd, 10-H), 5.80 (1H, ddd, 13-H), 6.09 (1H, br dd, 12-H), 6.59 (1H, dd, 11-H), 9.64 (1H, br s, 18-H).

9-*O*-(1-Ethoxyethyl)-3''-*O*-(methylthiomethyl)josamycin A_1 (**6b**)

Reaction of **5b** with MeOH gave **6b** as a colorless solid in 92% yield by a similar procedure to **6a**.

6b: MP 105~107°C; $[\alpha]_D^{18}$ −75° (*c* 1.0, MeOH); FD-MS *m/z* 959 (M^+); ^{1}H NMR δ 0.92 (1H, br ddd, 7-H), 0.98 (6H, d, 4''-OCOCH_2CH$(CH_3)_2$), 1.08 (3H, d, 6''-H), 1.14 (3H, br t, 9-OCH(OCH_2CH_3)CH_3), 1.15 (3H, d, 6'-H), 1.20 (3H, s, 3''-CH_3), 1.22 and 1.24 (3H, 2×d, 9-OCH(OCH_2CH_3)CH_3), 1.27 (3H, d, 16-H), 1.54 (1H, br dt, 7-H), 1.74 (1H, dd, 2''-Hax), 1.89 (1H, m, 8-H), 2.13 (1H, dt, 14-H), 2.20 (3H, s, 3''-OCH_2SCH_3), 2.26 (1H, br d, 2-H), 2.28 (3H, s, 3-OCOCH_3), 2.46 (1H, br dt, 14-H), 2.57 (6H, s, 3'-N$(CH_3)_2$), 2.75 and 2.76 (1H, 2×dd, 2-H), 2.88 (1H, br dd, 17-H), 3.58 (3H, s, 4-OCH_3), 3.63 (2H, q, 9-OCH(OCH_2CH_3)CH_3), 3.73 and 3.88 (1H, 2×dd, 9-H), 3.91 (1H, br d, 5-H), 4.51 (1H, d, 1'-H), 4.52 (1H, d, 3''-OCH_2SCH_3), 4.56 (1H, dq, 5''-H), 4.65, 4.66 (2H, 2×d, 4''-H, 3''-OCH_2SCH_3), 4.92 (1H, d, 1''-H), 5.03 (1H, ddq, 15-H), 5.11 (1H, br d, 3-H), 5.46 and 5.55 (1H, 2×dd, 10-H), 5.77 and 5.82 (1H, 2×ddd, 13-H), 6.09 (1H, br dd, 12-H), 6.56 and 6.60 (1H, 2×dd, 11-H), 9.64 and 9.65 (1H, 2×s, 18-H).

9-*O*-(1-Ethoxyethyl)-3''-*O*-methylmidecamycin A_1 (**7a**)

Raney-Nickel (7.5 ml) suspended in H_2O was washed with H_2O (12 ml) twice, and then washed with acetone (12 ml) three times below 25°C to be deactivated. Next, it was resuspended in EtOH (3.5 ml) twice to be used in this reaction.

To a solution of **6a** (300 mg, 0.317 mmol) in EtOH (12 ml) was added deactivated Raney-Nickel prepared above with EtOH (4.0 ml). The mixture was vigorously stirred at room temperature for 20 minutes exactly. Insoluble matter was filtered off, and it was washed with a mixed solvent (15 ml, EtOH-28% NH_4OH, 99:1) twice. Combined filtrate and washings were concentrated to give a residue which was purified with preparative TLC (toluene-acetone, 3:1) to afford **7a** (174 mg, 61%) as a colorless solid.

An isomer of **7a** derived from **5aH**: MP 90~93°C; $[\alpha]_D^{19}$ −43° (*c* 1.0, MeOH); SI-MS *m/z* 900 (MH^+); ^{1}H NMR δ 0.93 (1H, br ddd, 7-H), 0.98 (3H, d, 19-H), 1.08 (3H, d, 6''-H), 1.11 (3H, s, 3''-CH_3), 1.14 (3H, t, 9-OCH(OCH_2CH_3)CH_3), 1.16 (3H, d, 6'-H), 1.20 (3H, t, 4''-OCOCH_2CH_3), 1.22 (3H, d, 9-OCH(OCH_2CH_3)-CH_3), 1.23 (3H, t, 3-OCOCH_2CH_3), 1.26 (3H, d, 16-H), 1.54 (1H, br dt, 7-H), 1.67 (1H, dd, 2''-Hax), 1.89 (1H, m, 8-H), 2.13 (1H, br t, 6-H), 2.17 (1H, dt, 14-H), 2.26 (1H, br d, 2-H), 2.28 (1H, br dd, 17-H), 2.29 (1H, d, 2''-Heq), 2.43 and 2.44 (2H, 2×q, 4''-OCOCH_2CH_3), 2.50 and 2.64 (2H, 2×dq, 3-OCOCH_2CH_3), 2.60 (6H, s, 3'-N$(CH_3)_2$), 2.74 (1H, dd, 2-H), 2.85 (1H, br dd, 17-H), 3.26 (1H, br d, 4-H), 3.26 (3H, s, 3''-OCH_3), 3.29 (1H, dq, 5'-H), 3.43 and 3.50 (2H, 2×dq, 9-OCH(OCH_2-CH_3)CH_3), 3.46 (1H, t, 4'-H), 3.57 (3H, s, 4-OCH_3), 3.88 (1H, br d, 5-H), 3.90 (1H, dd, 9-H), 4.53 (1H, d, 1'-H), 4.54 (1H, dq, 5''-H), 4.64 (1H, q, 9-OC*H*(OCH_2CH_3)-CH_3), 4.73 (1H, d, 4''-H), 4.93 (1H, d, 1''-H), 5.00 (1H, ddq, 15-H), 5.13 (1H, br d, 3-H), 5.46 (1H, dd, 10-H), 5.83 (1H, ddd, 13-H), 6.10 (1H, br dd, 12-H), 6.62 (1H, dd, 11-H), 9.64 (1H, br s, 18-H).

An isomer of **7a** derived from **5aL**: MP 87~90°C; $[\alpha]_D^{19}$ −65° (*c* 1.0, MeOH); SI-MS *m/z* 900 (MH^+); ^{1}H NMR δ 0.94 (1H, br ddd, 7-H), 1.08 (3H, d, 6''-H), 1.10 (3H, s, 3''-CH_3), 1.14 (3H, t, 9-OCH(OCH_2CH_3)CH_3), 1.16 (3H, d, 6'-H), 1.18 (3H, t, 4''-OCOCH_2CH_3), 1.23 (3H, t, 3-OCOCH_2CH_3), 1.25 (3H, d, 9-OCH(OCH_2-CH_3)CH_3), 1.26 (3H, d, 16-H), 1.55 (1H, br dt, 7-H), 1.67 (1H, dd, 2''-Hax), 1.88 (1H, m, 8-H), 2.13 (1H, br t, 6-H), 2.16 (1H, dt, 14-H), 2.25 (1H, br dd, 17-H), 2.26 (1H, br d, 2-H), 2.29 (1H, d, 2''-Heq), 2.42 (1H, t, 3'-H), 2.43 and 2.44 (2H, 2×q, 4''-OCOCH_2CH_3), 2.52 and 2.64 (2H, 2×dq, 3-OCOCH_2CH_3), 2.57 (6H, s, 3'-N$(CH_3)_2$), 2.75 (1H, dd, 2-H), 2.86 (1H, br dd, 17-H), 3.21 (1H, dd, 2'-H), 3.26 (1H, br d, 4-H), 3.26 (3H, s, 3''-OCH_3), 3.29 (1H, dq, 5'-H), 3.36 and 3.63 (2H, 2×dq, 9-OCH(OCH_2CH_3)CH_3), 3.45 (1H, t, 4'-H), 3.57 (3H, s, 4-OCH_3), 3.75 (1H, dd, 9-H), 3.88 (1H, br d, 5-H), 4.52 (1H, d, 1'-H), 4.55 (1H, dq, 5''-H), 4.72 (1H, d, 4''-H), 4.92 (1H, d, 1''-H), 5.01 (1H, ddq, 15-H), 5.13 (1H, br d, 3-H), 5.55 (1H, dd, 10-H), 5.80 (1H, ddd, 13-H), 6.09 (1H, br dd, 12-H), 6.59 (1H, dd, 11-H), 9.64 (1H, br s, 18-H).

9-*O*-(1-Ethoxyethyl)-3''-*O*-methyljosamycin (**7b**)

Reaction of **6b** with Raney-Nickel gave **7b** as a colorless solid in 48% yield by a similar procedure to **7a**.

7b: MP 98~101°C; $[\alpha]_D^{16}$ −62° (*c* 1.0, MeOH); SI-MS *m/z* 914 (MH^+); ^1H NMR δ 0.97 (6H, d, 4″-$OCOCH_2CH(CH_3)_2$), 1.08 (3H, d, 6″-H), 1.10 (3H, s, 3″-CH_3), 1.14 (3H, br t, 9-$OCH(OCH_2CH_3)CH_3$), 1.16 (3H, d, 6′-H), 1.22 and 1.24 (3H, 2×d, 9-$OCH(OCH_2CH_3)CH_3$), 1.26 (3H, d, 16-H), 1.55 (1H, br dt, 7-H), 1.89 (1H, m, 8-H), 2.13 (1H, dt, 14-H), 2.28 (3H, s, 3-$OCOCH_3$), 2.46 (1H, br dt, 14-H), 2.58 (6H, s, 3′-$N(CH_3)_2$), 2.75 and 2.76 (1H, 2×dd, 2-H), 2.88 (1H, br dd, 17-H), 3.26 (3H, s, 3″-OCH_3), 3.35, 3.42, 3.50 and 3.63 (2H, 4×dq, 9-$OCH(OCH_2CH_3)CH_3$), 3.46 (1H, t, 4′-H), 3.58 (3H, s, 4-OCH_3), 3.73 and 3.88 (1H, 2×dd, 9-H), 3.91 (1H, br d, 5-H), 4.52 (1H, d, 1′-H), 4.54 (1H, dq, 5″-H), 4.63 and 4.64 (1H, 2×q, 9-$OCH(OCH_2CH_3)CH_3$), 4.73 (1H, d, 4″-H), 4.93 (1H, d, 1″-H), 5.03 (1H, ddq, 15-H), 5.12 (1H, br d, 3-H), 5.46 and 5.55 (1H, 2×dd, 10-H), 5.77 and 5.82 (1H, 2×ddd, 13-H), 6.09 (1H, br dd, 12-H), 6.57 and 6.59 (1H, 2×dd, 11-H), 9.64 and 9.66 (1H, 2×s, 18-H).

3″-*O*-Methylmidecamycin A_1 (**8a**)

To a solution of **7a** (60.0 mg, 6.67×10^{-5} mol) in CH_3CN (1.5 ml) was added 5% (v/v) aqueous acetic acid (4.5 ml). The solution was allowed to stand at room temperature for 16 hours. Evaporation gave a residue which was extracted with $CHCl_3$ (10 ml). The organic layer was washed with saturated aqueous $NaHCO_3$ (10 ml) three times, brine (10 ml) and dried. Concentration gave a residue which was purified by preparative TLC ($CHCl_3$ - MeOH, 10 : 1) to afford **8a** (50.0 mg, 91%) as a colorless solid: MP 116~120°C; $[\alpha]_D^{15}$ −65° (*c* 1.0, MeOH); SI-MS *m/z* 828 (MH^+); ^1H NMR δ 0.92 (1H, br ddd, 7-H), 0.98 (3H, d, 19-H), 1.07 (3H, d, 6″-H), 1.10 (3H, s, 3″-CH_3), 1.16 (3H, d, 6′-H), 1.17 (3H, t, 4″-$OCOCH_2CH_3$), 1.22 (3H, t, 3-$OCOCH_2CH_3$), 1.26 (3H, d, 16-H), 1.54 (1H, br dt, 7-H), 1.66 (1H, dd, 2″-Hax), 1.89 (1H, m, 8-H), 2.15 (1H, dt, 14-H), 2.24 (1H, br d, 2-H), 2.29 (1H, d, 2″-Heq), 2.32 (1H, br dd, 17-H), 2.42 (1H, t, 3′-H), 2.42 and 2.43 (2H, 2×q, 4″-$OCOCH_2CH_3$), 2.51 and 2.64 (2H, 2×dq, 3-$OCOCH_2CH_3$), 2.57 (6H, s, 3′-$N(CH_3)_2$), 2.76 (1H, dd, 2-H), 2.86 (1H, br dd, 17-H), 3.22 (1H, dd, 2′-H), 3.26 (1H, br d, 4-H), 3.26 (3H, s, 3″-OCH_3), 3.29 (1H, dq, 5′-H), 3.44 (1H, t, 4′-H), 3.57 (3H, s, 4-OCH_3), 3.88 (1H, br d, 5-H), 4.07 (1H, dd, 9-H), 4.52 (1H, d, 1′-H), 4.54 (1H, dq, 5″-H), 4.72 (1H, d, 4″-H), 4.92 (1H, d, 1″-H), 5.03 (1H, ddq, 15-H), 5.14 (1H, br d, 3-H), 5.61 (1H, dd, 10-H), 5.79 (1H, ddd, 13-H), 6.08 (1H, br dd, 12-H), 6.67 (1H, dd, 11-H), 9.63 (1H, br s, 18-H).

3″-*O*-Methyljosamycin (**8b**)

Reaction of **7b** with acetic acid gave **8b** as a colorless solid in 84% yield by a similar procedure to **8a**.

8b: MP 115~117°C; $[\alpha]_D^{17}$ −65° (*c* 1.0, MeOH); SI-MS *m/z* 842 (MH^+); ^1H NMR δ 0.92 (1H, br ddd, 7-H), 0.97 (6H, d, 4″-$OCOCH_2CH(CH_3)_2$), 0.98 (3H, d, 19-H), 1.08 (3H, d, 6″-H), 1.10 (3H, s, 3″-CH_3), 1.16 (3H, d, 6′-H), 1.26 (3H, d, 16-H), 1.58 (1H, br dt, 7-H), 1.66 (1H, dd, 2″-Hax), 1.89 (1H, m, 8-H), 2.28 (3H, s, 3-$OCOCH_3$), 2.33 (1H, br dd, 17-H), 2.41 (1H, t, 3′-H), 2.46 (1H, br dt, 14-H), 2.57 (6H, s, 3′-$N(CH_3)_2$), 2.74 (1H, dd, 2-H), 2.88 (1H, br dd, 17-H), 3.21 (1H, dd, 2′-H), 3.25 (3H, s, 3″-OCH_3), 3.26 (1H, br d, 4-H), 3.29 (1H, dq, 5′-H), 3.46 (1H, t, 4′-H), 3.58 (3H, s, 4-OCH_3), 3.90 (1H, br d, 5-H), 4.05 (1H, dd, 9-H), 4.52 (1H, d, 1′-H), 4.72 (1H, d, 4″-H), 4.93 (1H, br d, 1″-H), 5.04 (1H, ddq, 15-H), 5.12 (1H, br d, 3-H), 5.54 (1H, dq, 5″-H), 5.62 (1H, dd, 10-H), 5.76 (1H, ddd, 13-H), 6.08 (1H, br dd, 12-H), 6.64 (1H, dd, 11-H), 9.64 (1H, s, 18-H).

9-*O*-Acetyl-3″-*O*-methylmidecamycin A_1 (**9a**) from **8a**

To a solution of **8a** (60.0 mg, 7.25×10^{-5} mol) in toluene (3.0 ml) was successively added anhydrous pyridine (26 ml, 0.32 mmol) and acetyl chloride (23 ml, 0.32 mmol). After stirring at room temperature for one hour, Et_3N (19 ml, 0.27 mmol) and EtOAc (30 ml) was added. The organic layer was washed with H_2O (30 ml) twice, dried and concentrated to give a residue which was purified by preparative TLC ($CHCl_3$ - MeOH, 12 : 1) to afford **9a** (24.9 mg, 79%) as colorless needles: MP 118~121°C; $[\alpha]_D^{24}$ −60° (*c* 1.0, MeOH); EI-MS *m/z* 869 (M^+); ^1H NMR δ 0.93 (1H, br ddd, 7-H), 0.96 (3H, d, 19-H), 1.08 (3H, d, 6″-H), 1.10 (3H, s, 3″-CH_3), 1.16 (3H, d, 6′-H), 1.18 (3H, t, 4″-$OCOCH_2CH_3$), 1.21 (3H, t, 3-$OCOCH_2CH_3$), 1.26 (3H, d, 16-H), 1.57 (1H, br dt, 7-H), 1.67 (1H, dd, 2″-Hax), 2.02 (3H, s, 9-$OCOCH_3$), 2.17 (1H, dt, 14-H), 2.26 (1H, br d, 2-H), 2.30 (1H, d, 2″-Heq), 2.41 (1H, t, 3′-H), 2.43 and 2.44 (2H, 2×q, 4″-$OCOCH_2CH_3$), 2.51 and 2.67 (2H, 2×dq, 3-$OCOCH_2CH_3$), 2.57 (6H, s, 3′-$N(CH_3)_2$), 2.58 (1H, br dd, 17-H), 2.74 (1H, dd, 2-H), 2.83 (1H, br dd, 17-H), 3.20 (1H, dd, 2′-H), 3.25 (1H, br d, 4-H), 3.26 (3H, s, 3″-OCH_3), 3.28 (1H, dq, 5′-H), 3.45 (1H, t, 4′-H), 3.57 (3H, s, 4-OCH_3), 3.93 (1H, br d, 5-H), 4.51 (1H, d, 1′-H), 4.54 (1H, dq, 5″-H), 4.72 (1H, d, 4″-H), 4.93 (1H, d, 1″-H), 4.98 (1H, ddq, 15-H), 5.08 (1H, dd, 9-H), 5.12 (1H, br d, 3-H), 5.57 (1H, dd, 10-H), 5.88 (1H, ddd, 13-H), 6.09 (1H, br dd, 12-H), 6.74 (1H, dd, 11-H), 9.65 (1H, br s, 18-H).

3″-*O*-Methyl-9-*O*-propionylmidecamycin A_1 (**10a**) from **8a**

Reaction of **8a** with propionyl chloride gave **10a** as a colorless solid in 78% yield by a similar procedure to **9a** from **8a**.

10a: MP 114~117°C; $[\alpha]_D^{22}$ −72° (*c* 1.0, MeOH); EI-MS *m/z* 883 (M^+); ^1H NMR δ 0.90 (1H, br ddd, 7-H), 0.96 (3H, d, 19-H), 1.08 (3H, d, 6″-H), 1.10 (3H, s, 3″-CH_3), 1.11 (3H, t, 9-$OCOCH_2CH_3$), 1.16 (3H, d, 6′-H), 1.17 (3H, t, 4″-$OCOCH_2CH_3$), 1.21 (3H, t, 3-$OCOCH_2CH_3$), 1.26 (3H, d, 16-H), 1.57 (1H, br dt, 7-H), 1.67 (1H, dd, 2″-Hax), 2.02 (1H, m, 8-H), 2.17 (1H, dt, 14-H), 2.26 (1H, br d, 2-H), 2.29 (1H, d, 2″-Heq), 2.30 (2H, q, 9-$OCOCH_2CH_3$), 2.43 and 2.44 (2H, 2×q, 4″-$OCOCH_2CH_3$), 2.51 and 2.68 (2H, 2×dq, 3-$OCOCH_2CH_3$), 2.58 (6H, s, 3′-$N(CH_3)_2$), 2.60 (1H, br dd, 17-H), 2.74 (1H, dd, 2-H), 2.83 (1H, br dd, 17-H), 3.21 (1H, dd, 2′-H), 3.24 (1H, br d, 4-H), 3.28 (1H, dq, 5′-H), 3.26 (3H, s, 3″-OCH$_3$), 3.45 (1H, t, 4′-H), 3.56 (3H, s, 4-OCH$_3$), 3.94 (1H, br d, 5-H), 4.51 (1H, d, 1′-H), 4.53 (1H, dq, 5″-H), 4.72 (1H, d, 4″-H), 4.93 (1H, d, 1″-H), 4.98 (1H, ddq, 15-H), 5.09 (1H, dd, 9-H), 5.12 (1H, br d, 3-H), 5.58 (1H, dd, 10-H), 5.88 (1H, ddd, 13-H), 6.09 (1H, br dd, 12-H), 6.74 (1H, dd, 11-H), 9.65 (1H, br s, 18-H).

9,2′-Di-*O*-acetyl-3″-*O*-(methylthiomethyl)josamycin (**13b**)

Reaction of **11b**[22] with DMSO gave **13b** as a colorless solid in 66% yield by a similar procedure to **5a**.

13b: MP 118~122°C; $[\alpha]_D^{24}$ −85° (*c* 1.0, CHCl$_3$); SI-MS *m/z* 972 (MH$^+$); ^{1}H NMR δ 0.85 (1H, br ddd, 7-H), 0.96 (3H, d, 19-H), 0.98 (6H, d, 4″-OCOCH$_2$CH-(C*H*$_3$)$_2$), 1.05 (3H, d, 6″-H), 1.14 (3H, d, 6′-H), 1.18 (3H, s, 3″-CH$_3$), 1.26 (3H, d, 16-H), 1.46 (1H, br dt, 7-H), 1.68 (1H, dd, 2″-Hax), 2.00 (3H, s, 9-OCOCH$_3$), 2.00 (3H, s, 2′-OCOCH$_3$), 2.20 (3H, s, 3″-OCH$_2$SC*H*$_3$), 2.25 (1H, br d, 2-H), 2.30 (3H, s, 3-OCOCH$_3$), 2.42 (6H, s, 3′-N(CH$_3$)$_2$), 2.45 (1H, br dt, 14-H), 2.55 (1H, br dd, 17-H), 2.68 (1H, t, 3′-H), 2.72 (1H, dd, 2-H), 2.81 (1H, br d, 17-H), 3.16 (1H, br d, 4-H), 3.16 (1H, t, 4′-H), 3.26 (1H, dq, 5′-H), 3.50 (3H, s, 4-OCH$_3$), 3.94 (1H, br d, 5-H), 4.50 and 4.64 (2H, 2×d, 3″-OC*H*$_2$SCH$_3$), 4.56 (1H, dq, 5″-H), 4.59 (1H, d, 1′-H), 4.63 (1H, d, 4″-H), 4.81 (1H, d, 1″-H), 4.91 (1H, dd, 2′-H), 4.99 (1H, ddq, 15-H), 5.05 (1H, dd, 9-H), 5.09 (1H, br d, 3-H), 5.56 (1H, dd, 10-H), 5.85 (1H, ddd, 13-H), 6.05 (1H, br dd, 12-H), 6.70 (1H, dd, 11-H), 9.63 (1H, br s, 18-H).

2′-*O*-Acetyl-3″-*O*-(methylthiomethyl)-9-*O*-propionylmidecamycin A$_1$ (**14a**)

Reaction of **12a** with DMSO gave **14a** as a colorless solid in 57% yield by a similar procedure to **5a**.

14a: MP 97~100°C; $[\alpha]_D^{28}$ −81° (*c* 1.0, CHCl$_3$); FAB-MS *m/z* 930 (MH$^+$); ^{1}H NMR δ 0.86 (1H, br ddd, 7-H), 0.96 (3H, d, 19-H), 1.05 (3H, d, 6″-H), 1.11 (3H, t, 9-OCOCH$_2$C*H*$_3$), 1.14 (3H, d, 6′-H), 1.17 (3H, s, 3″-CH$_3$), 1.18 (3H, t, 4″-OCOCH$_2$C*H*$_3$), 1.21(3H, t, 3-OCOCH$_2$C*H*$_3$), 1.26 (3H, d, 16-H), 1.45 (1H, br dt, 7-H), 1.68 (1H, dd, 2″-Hax), 2.01 (3H, s, 2′-OCOCH$_3$), 2.16 (1H, dt, 14-H), 2.20 (3H, s, 3″-OCH$_2$SC*H*$_3$), 2.24 (1H, br d, 2-H), 2.26 (1H, d, 2″-Heq), 2.29 (2H, q, 9-OCOC*H*$_2$CH$_3$), 2.42 (2H, q, 4″-OCOC*H*$_2$CH$_3$), 2.42 (6H, s, 3′-N(CH$_3$)$_2$), 2.51 and 2.69 (2H, 2×dq, 3-OCOC*H*$_2$CH$_3$), 2.56 (1H, br dd, 17-H), 2.71 (1H, dd, 2-H), 2.80 (1H, br dd, 17-H), 3.15 (1H, br dd, 4-H), 3.15 (1H, t, 4′-H), 3.26 (1H, dq, 5′-H), 3.49 (3H, s, 4-OCH$_3$), 3.92 (1H, br d, 5-H), 4.51 (1H, d, 3″-OC*H*$_2$SCH$_3$), 4.56 (1H, dq, 5″-H), 4.59 (1H, d, 1′-H), 4.63 (1H, d, 4″-H), 4.65 (1H, d, 3″-OC*H*$_2$SCH$_3$), 4.81 (1H, d, 1″-H), 4.90 (1H, dd, 2′-H), 4.96 (1H, ddq, 15-H), 5.07 (1H, dd, 9-H), 5.10 (1H, br d, 3-H), 5.57 (1H, dd, 10-H), 5.88 (1H, ddd, 13-H), 6.05 (1H, br dd, 12-H), 6.12 (1H, dd, 11-H), 9.63 (1H, br s, 18-H).

2′-*O*-Acetyl-3″-*O*-(methylthiomethyl)-9-*O*-propionyljosamycin (**14b**)

Reaction of **12b**[23] with DMSO gave **14b** as a colorless solid in 47% yield by a similar procedure to **5a**.

14b: MP 114~116°C; $[\alpha]_D^{16}$ −90° (*c* 1.0, CHCl$_3$); SI-MS *m/z* 986 (MH$^+$); ^{1}H NMR δ 0.85 (1H, br dt, 7-H), 0.95 (3H, d, 19-H), 0.98 (6H, d, 4″-OCOCH$_2$CH(C*H*$_3$)$_2$), 1.05 (3H, d, 6″-H), 1.10 (3H, t, 9-OCOCH$_2$C*H*$_3$), 1.14 (3H, d, 6′-H), 1.18 (3H, s, 3″-CH$_3$), 1.26 (3H, d, 16-H), 1.46 (1H, br dt, 7-H), 1.68 (1H, dd, 2″-Hax), 2.01 (3H, s, 2′-OCOCH$_3$), 2.20 (3H, s, 3″-OCH$_2$SC*H*$_3$), 2.25 (1H, br d, 2-H), 2.30 (3H, s, 3-OCOCH$_3$), 2.42 (6H, s, 3′-N(CH$_3$)$_2$), 2.45 (1H, br dt, 14-H), 2.58 (1H, br dd, 17-H), 2.67 (1H, t, 3′-H), 2.71 (1H, dd, 2-H), 2.82 (1H, br dd, 17-H), 3.16 (1H, br d, 4-H), 3.16 (1H, t, 4′-H), 3.26 (1H, dq, 5′-H), 3.49 (3H, s, 4-OCH$_3$), 3.94 (1H, br d, 5-H), 4.50 (1H, d, 3″-OC*H*$_2$SCH$_3$), 4.56 (1H, dq, 5″-H), 4.59 (1H, d, 1′-H), 4.63, 4.64 (2H, 2×d, 4″-H, 3″-OC*H*$_2$SCH$_3$), 4.81 (1H, d, 1″-H), 4.90 (1H, dd, 2′-H), 4.98 (1H, ddq, 15-H), 5.05 (1H, dd, 9-H), 5.09 (1H, br d, 3-H), 5.57 (1H, dd, 10-H), 5.85 (1H, ddd, 13-H), 6.05 (1H, br dd, 12-H), 6.69 (1H, dd, 11-H), 9.64 (1H, s, 18-H).

9-*O*-Acetyl-3″-*O*-(methylthiomethyl)josamycin (**15b**)

Reaction of **13b** with MeOH gave **15b** as a colorless solid in 88% yield by a similar procedure to **6a**.

15b: MP 115~118°C; $[\alpha]_D^{24}$ −77° (*c* 1.0, MeOH); SI-MS *m/z* 930 (MH$^+$); ^{1}H NMR δ 0.93 (1H, br ddd, 7-H), 0.96 (3H, d, 19-H), 0.98 (6H, d, 4″-OCOCH$_2$CH-(C*H*$_3$)$_2$), 1.08 (3H, d, 6″-H), 1.15 (3H, d, 6′-H), 1.20 (3H, s, 3″-CH$_3$), 1.27 (3H, d, 16-H), 1.57 (1H, br dt, 7-H), 1.74 (1H, dd, 2″-Hax), 2.01 (3H, s, 9-OCOCH$_3$), 2.18 (3H, s, 3″-OCH$_2$SC*H*$_3$), 2.27 (1H, br d, 2-H), 2.29 (3H, s, 3-OCOCH$_3$), 2.42 (1H, t, 3′-H), 2.47 (1H, br dt, 14-H), 2.58 (6H, s, 3′-N(CH$_3$)$_2$), 2.59 (1H, br dd, 17-H), 2.75 (1H, dd, 2-H), 2.84 (1H, br dd, 17-H), 3.21 (1H, dd, 2′-H), 3.25 (1H, br d, 4-H), 3.28 (1H, dq, 5′-H), 3.42 (1H, t, 4′-H), 3.57 (3H, s, 4-OCH$_3$), 3.96 (1H, br d, 5-H), 4.50 (1H, d, 1′-H), 4.52 and 4.65 (2H, 2×d, 3″-OC*H*$_2$SCH$_3$), 4.56 (1H, dq, 5″-H), 4.66 (1H, d, 4″-H), 4.92 (1H, d, 1″-H), 4.97 (1H, dd, 9-H), 5.00 (1H, br d, 3-H), 5.00 (1H, ddq, 15-H), 5.57 (1H, dd, 10-H), 5.86 (1H, ddd, 13-H), 6.09 (1H, br dd, 12-H), 6.71 (1H, dd, 11-H), 9.66 (1H, br s, 18-H).

3″-*O*-(Methylthiomethyl)-9-*O*-propionylmidecamycin A$_1$ (**16a**)

Reaction of **14a** with MeOH gave **16a** as a colorless solid in 96% yield by a similar procedure to **6a**.

16a: MP 113~117°C; $[\alpha]_D^{28}$ −66° (*c* 1.0, MeOH); FAB-MS *m/z* 972 (MH$^+$); ^{1}H NMR δ 0.94 (1H, br ddd, 7-H), 0.95 (3H, d, 19-H), 1.08 (3H, d, 6″-H), 1.11 (3H, t, 9-OCOCH$_2$C*H*$_3$), 1.15 (3H, d, 6′-H), 1.19 (3H, t, 4″-OCOCH$_2$C*H*$_3$), 1.20 (3H, s, 3″-CH$_3$), 1.21 (3H, t, 3-OCOCH$_2$C*H*$_3$), 1.26 (3H, d, 16-H), 1.56 (1H, br dt, 7-H), 1.75 (1H, dd, 2″-Hax), 2.17 (1H, dt, 14-H), 2.19 (3H, s, 3″-OCH$_2$SC*H*$_3$), 2.26 (1H, br d, 2-H), 2.28 (1H, d, 2″-Heq), 2.29 (2H, q, 9-OCOC*H*$_2$CH$_3$), 2.42 (2H, q, 4″-OCOCH_2CH_3), 2.51 and 2.68 (2H, 2×dq, 3-OCOCH_2CH_3), 2.58 (6H, s, 3′-N(CH_3)$_2$), 2.61 (1H, br dd, 17-H), 2.74 (1H, dd, 2-H), 2.82 (1H, br dd, 17-H), 3.22 (1H, dd, 2′-H), 3.25 (1H, br dd, 4-H), 3.27 (1H, dq, 5′-H), 3.41 (1H, t, 4′-H), 3.56 (3H, s, 4-OCH_3), 3.93 (1H, br d, 5-H), 4.49 (1H, d, 1′-H), 4.52 (1H, d, 3″-OCH_2SCH_3), 4.55 (1H, dq, 5″-H), 4.65 (1H, d, 3″-OCH_2SCH_3), 4.66 (1H, d, 4″-H), 4.92 (1H, d, 1″-H), 4.98 (1H, ddq, 15-H), 5.09 (1H, dd, 9-H), 5.12 (1H, br d, 3-H), 5.58 (1H, dd, 10-H), 5.88 (1H, ddd, 13-H), 6.09 (1H, br dd, 12-H), 6.74 (1H, dd, 11-H), 9.65 (1H, br s, 18-H).

3″-*O*-(Methylthiomethyl)-9-*O*-propionyljosamycin (**16b**)

Reaction of **14b** with MeOH gave **16b** as a colorless solid in 92% yield by a similar procedure to **6a**.

16b: MP 113~116°C; $[\alpha]_D^{16}$ −58° (*c* 1.0, MeOH); SI-MS *m/z* 944 (MH$^+$); ^{1}H NMR δ 0.92 (1H, br dt, 7-H), 0.95 (3H, d, 19-H), 0.98 (6H, d, 4″-OCOCH_2CH(CH_3)$_2$), 1.08 (3H, d, 6″-H), 1.10 (3H, t, 9-OCOCH_2CH_3), 1.15 (3H, d, 6′-H), 1.20 (3H, s, 3″-CH_3), 1.26 (3H, d, 16-H), 1.57 (1H, br dt, 7-H), 1.74 (1H, dd, 2″-Hax), 2.03 (1H, m, 8-H), 2.19 (3H, s, 3″-OCH_2SCH_3), 2.26 (1H, br d, 2-H), 2.29 (3H, s, 3-OCOCH_3), 2.42 (1H, t, 3′-H), 2.46 (1H, br dt, 14-H), 2.57 (6H, s, 3′-N(CH_3)$_2$), 2.62 (1H, br dd, 17-H), 2.75 (1H, dd, 2-H), 2.84 (1H, br dd, 17-H), 3.21 (1H, dd, 2′-H), 3.25 (1H, br d, 4-H), 3.28 (1H, dq, 5′-H), 3.42 (1H, t, 4′-H), 3.57 (3H, s, 4-OCH_3), 3.96 (1H, br d, 5-H), 4.49 (1H, d, 1′-H), 4.52 (1H, d, 3″-OCH_2SCH_3), 4.55 (1H, dq, 5″-H), 4.64, 4.66 (2H, 2×d, 4″-H, 3″-OCH_2SCH_3), 4.92 (1H, d, 1″-H), 5.00 (1H, ddq, 15-H), 5.07 (1H, dd, 9-H), 5.10 (1H, br d, 3-H), 5.58 (1H, dd, 10-H), 5.86 (1H, ddd, 13-H), 6.09 (1H, br dd, 12-H), 6.71 (1H, dd, 11-H), 9.66 (1H, s, 18-H).

9-*O*-Acetyl-3″-*O*-methylmidecamycin A_1 (**9a**) from **15a**

Reaction of **15a**[14] with Raney-Nickel gave **9a** as colorless needles in 67% yield by a similar procedure to **7a**.

9-*O*-Acetyl-3″-*O*-methyljosamycin (**9b**)

Reaction of **15b** with Raney-Nickel gave **9b** as a colorless solid in 53% yield by a similar procedure to **7a**.

9b: MP 115~119°C; $[\alpha]_D^{26}$ −74° (*c* 1.0, MeOH); SI-MS *m/z* 884 (MH$^+$); ^{1}H NMR δ 0.92 (1H, br ddd, 7-H), 0.96 (3H, d, 19-H), 0.97 (6H, d, 4″-OCOCH_2CH(CH_3)$_2$), 1.08 (3H, d, 6″-H), 1.11 (3H, s, 3″-CH_3), 1.16 (3H, d, 6′-H), 1.26 (3H, d, 16-H), 1.58 (1H, br dt, 7-H), 1.67 (1H, dd, 2″-Hax), 2.01 (3H, s, 9-OCOCH_3), 2.29 (3H, s, 3-OCOCH_3), 2.42 (1H, t, 3′-H), 2.58 (6H, s, 3′-N(CH_3)$_2$), 2.59 (1H, br dd, 17-H), 2.75 (1H, dd, 2-H), 2.84 (1H, br dd, 17-H), 3.19 (1H, dd, 2′-H), 3.25 (1H, br d, 4-H), 3.26 (3H, s, 3″-OCH_3), 3.29 (1H, dq, 5′-H), 3.46 (1H, t, 4′-H), 3.57 (3H, s, 4-OCH_3), 3.96 (1H, br d, 5-H), 4.51 (1H, d, 1′-H), 4.54 (1H, dq, 5″-H), 4.72 (1H, d, 4″-H), 4.93 (1H, d, 1″-H), 4.99 (1H, ddq, 15-H), 5.06 (1H, dd, 9-H), 5.12 (1H, br d, 3-H), 5.57 (1H, dd, 10-H), 5.86 (1H, ddd, 13-H), 6.09 (1H, br dd, 12-H), 6.71 (1H, dd, 11-H), 9.66 (1H, br s, 18-H).

3″-*O*-Methyl-9-*O*-propionylmidecamycin A_1 (**10a**) from **16a**

Reaction of **16a** with Raney-Nickel gave **10a** as a colorless solid in 35% yield by a similar procedure to **7a**.

3″-*O*-Methyl-9-*O*-propionyljosamycin (**10b**)

Reaction of **16b** with Raney-Nickel gave **10b** as a colorless solid in 39% yield by a similar procedure to **7a**.

10b: MP 115~118°C; $[\alpha]_D^{15}$ −61° (*c* 1.0, MeOH); SI-MS *m/z* 898 (MH$^+$); ^{1}H NMR δ 0.92 (1H, br dt, 7-H), 0.95 (3H, d, 19-H), 0.97 (6H, d, 4″-OCOCH_2CH(CH_3)$_2$), 1.08 (3H, d, 6″-H), 1.11 (3H, t, 9-OCOCH_2CH_3), 1.11 (3H, s, 3″-CH_3), 1.16 (3H, d, 6′-H), 1.26 (3H, d, 16-H), 1.58 (1H, br dt, 7-H), 1.67 (1H, dd, 2″-Hax), 2.03 (1H, m, 8-H), 2.17 (1H, dt, 14-H), 2.26 (1H, br d, 2-H), 2.29 (3H, s, 3-OCOCH_3), 2.41 (1H, t, 3′-H), 2.46 (1H, br dt, 14-H), 2.57 (6H, s, 3′-N(CH_3)$_2$), 2.62 (1H, br dd, 17-H), 2.75 (1H, dd, 2-H), 2.85 (1H, br dd, 17-H), 3.19 (1H, dd, 2′-H), 3.24 (1H, br d, 4-H), 3.26 (3H, s, 3″-OCH_3), 3.29 (1H, dq, 5′-H), 3.45 (1H, t, 4′-H), 3.58 (3H, s, 4-OCH_3), 3.96 (1H, br d, 5-H), 4.51 (1H, d, 1′-H), 4.54 (1H, dq, 5″-H), 4.73 (1H, d, 4″-H), 4.93 (1H, d, 1″-H), 5.00 (1H, ddq, 15-H), 5.07 (1H, dd, 9-H), 5.11 (1H, br d, 3-H), 5.58 (1H, dd, 10-H), 5.86 (1H, ddd, 13-H), 6.09 (1H, br dd, 12-H), 6.71 (1H, dd, 11-H), 9.66 (1H, s, 18-H).

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