TOTAL SYNTHESIS OF 3-*O*-DEMETHYLSPORARICIN A

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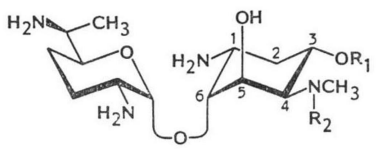
The novel, semisynthetic pseudodisaccharide antibiotic, 3-*O*-demethylsporaricin A, was synthesized *via* 3-*O*-demethylsporaricin B obtained by glycosidation of its aminocyclitol part. The aminocyclitol part was synthesized as D,L-form from D,L-(1,2,3/4,5,6)-1,4-bis(benzyloxycarbonylamino)-5,6-*O*-isopropylidene-2,3,5,6-cyclohexanetetraol *via* three key steps, namely, deoxygenation, inversion of a hydroxyl group, and *N*-methylation. The physical and biological properties of synthetic 3-*O*-demethylsporaricin A and an authentic sample derived from sporaricin B were identical.

The novel pseudodisaccharide antibiotics, sporaricin A and sporaricin B, have been isolated from the culture broth of *Saccharopolyspora hirsuta* subsp. *kobensis* by DEUSHI *et al.*[1,2]. The structures of sporaricin A and sporaricin B have been determined as shown in Fig. 1 by chemical degradation[3]. Sporaricin A possesses superior activity against Gram-positive and Gram-negative bacteria including traditional aminoglycoside-resistant strains.

Recently, 3-*O*-demethylsporaricin A (**1**) has been derived from sporaricin B[4]. It exhibits stronger antibacterial activity than sporaricin A especially against *Pseudomonas aeruginosa*[5]. Nowadays, *P. aeruginosa* is one of the most important pathogenic microorganism. Thus, we became much interested in the antibacterial activity of **1** and planned its total synthesis.

In relation to the total synthesis of 3-*O*-demethylsporaricins, we have reported the synthesis of its aminocyclitol part as D,L-form[6] and the synthesis of its sugar part, 6-*epi*-purpurosamine B[7]. The present report describes total synthesis of 3-*O*-demethylsporaricin A.

Fig. 1. Sporaricins.



	R_1	R_2
Sporaricin A	CH_3	Gly
Sporaricin B	CH_3	H
3-*O*-Demethylsporaricin A (**1**)	H	Gly
3-*O*-Demethylsporaricin B (**34**)	H	H

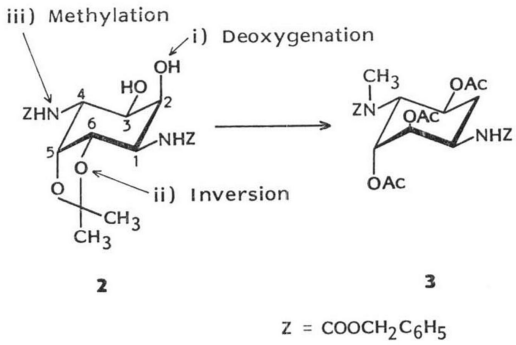
Gly: $COCH_2NH_2$.

Synthesis of the Aminocyclitol Part of 3-*O*-Demethylsporaricin A

The sporaricins possess a 1,4-*trans*-diaminocyclitol moiety in contrast with other pseudodisaccharide antibiotics such as fortimicins[8], istamycin A[9], and lysinomicin[10], which are glycosides of a 1,4-*cis*-diaminocyclitol. Therefore, we selected a 1,4-*trans*-diaminocyclitol derivative, D,L-(1,2,3/4,5,6)-1,4-bis(benzyloxycarbonylamino)-5,6-*O*-isopropylidene-2,3,5,6-cyclohexanetetraol† (**2**), as a starting material for the synthesis of D,L-(1,3,6/4,5)-3,5,6-tri-*O*-acetyl-1,4-bis(benzyloxycarbonylamino)-4-*N*-methyl-3,5,6-cyclohexanetriol (**3**), namely, a protected D,L-form of the aminocyclitol part of 3-*O*-

† The trivial numbering system of sporaricins is employed in order to avoid confusion (Fig. 1)[5].

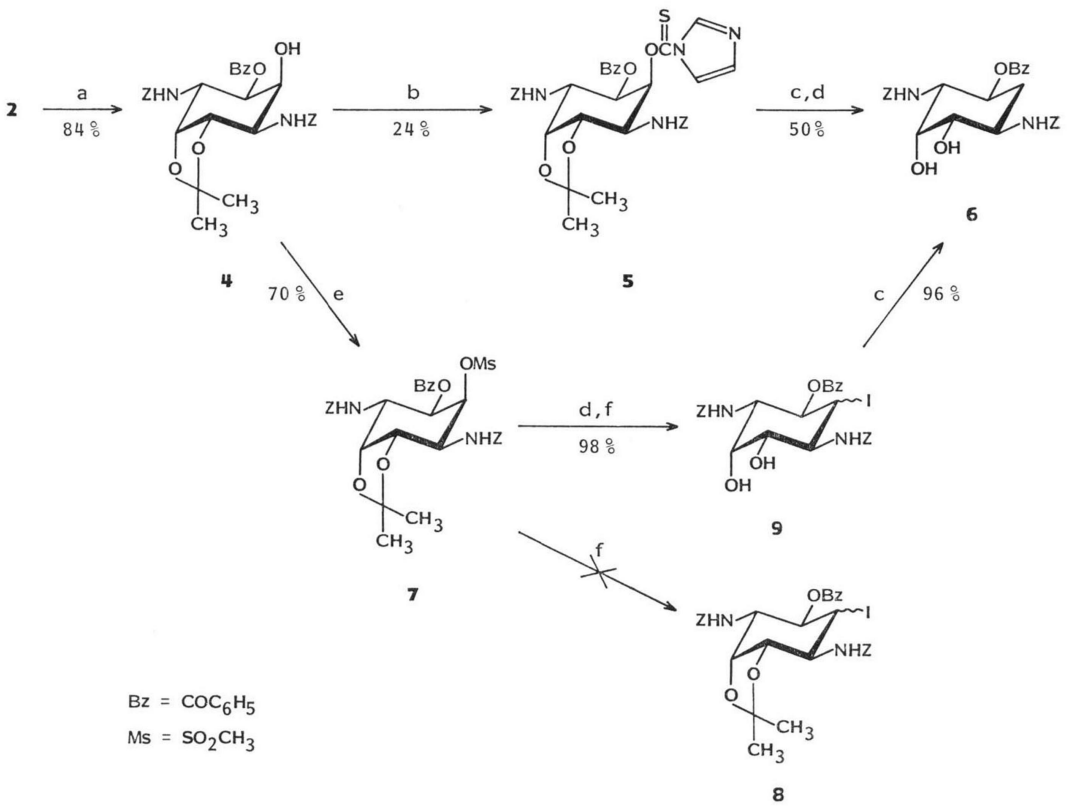
Scheme 1. Our synthetic strategy for the aminocyclitol part of 3-*O*-demethylsporaricin A.



demethylsporaricin A. The starting compound **2** was easily obtained from nitromethane and glyoxal in four steps[11]. In order to transform **2** into **3**, three key steps are required; i) deoxygenation at the 2-position, ii) inversion of the 6-hydroxyl group, and iii) methylation of the 4-amino group (Scheme 1).

The first step required deoxygenation of the 2-position in **2**. Regioselective protection of the equatorial hydroxyl group in **2** gave the corresponding 3-*O*-benzoyl derivative (**4**) in 84% yield. Treatment of **4** with 1,1′-thiocarbonyldiimidazole in tetrahydrofuran afforded the corresponding 2-*O*-imidazolylthiocarbonyl derivative (**5**) in 24% yield. Reduction of **5** with tri-*n*-butyltin hydride in the presence of 2,2′-azobisisobutyronitrile in toluene, followed by removal of the isopropylidene group by 80% acetic acid, gave D,L-(1,3/4,5,6)-3-*O*-benzoyl-1,4-bis(benzyloxycarbonylamino)-3,5,6-cyclohexanetriol (**6**) in 50% yield. Several attempts to improve the yield of **5** were unsuccessful. Therefore, an alternative route was investigated. Mesyla-

Scheme 2. Deoxygenation step.

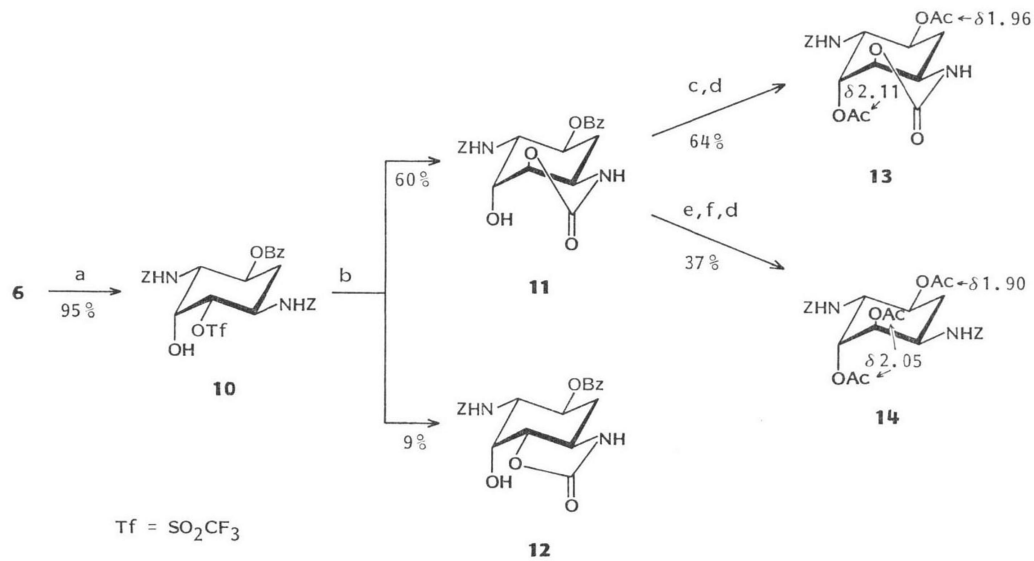


a: BzCl - pyridine, b: thiocarbonyldiimidazole - THF, c: $(n\text{-Bu})_3SnH$ - 2,2′-azobisisobutyronitrile, d: AcOH - H_2O, 4: 1, e: MsCl - pyridine, f: NaI - DMF.

tion of **4** gave the corresponding 2-*O*-mesyl derivative (**7**) in 70% yield. But, treatment of **7** with excess sodium iodide in *N*,*N*-dimethylformamide did not afford the corresponding 2-iodo derivative (**8**). We assumed that the bicyclic structure of **7** was so rigid that S_N-2 attack by iodide ion did not proceed. Accordingly, removal of the isopropylidene group in **7** must be necessary before the substitution by iodide ion. Treatment of **7** with 80% acetic acid, followed by treatment with excess sodium iodide in *N*,*N*-dimethylformamide at 100°C for 8 hours, gave the corresponding 2-iodo-5,6-diol derivative (**9**)* in 98% yield. Reduction of **9** with tri-*n*-butyltin hydride in the presence of 2,2′-azobisisobutyronitrile afforded **6** in 96% yield. Thus, the overall yield of **6** from **4** was increased from 12% to 66%.

The next step required inversion of the 6-hydroxyl group in **6**. We planned to use the neighboring effect of the benzyloxycarbonylamino group at the 1-position in **6**, and considered that a *cis*-cyclic carbamate could be formed by converting the 6-hydroxyl group to a good leaving group[12]. For this purpose, the trifluoromethanesulfonyl group was selected as the leaving group. Regioselective trifluoromethanesulfonylation of the equatorial hydroxyl group in **6** gave the corresponding 6-*O*-trifluoromethanesulfonyl derivative (**10**) in 95% yield. Heating **10** at 60~65°C in *N*,*N*-dimethylformamide for 2.5 hours afforded D,L-(1,3,6/4,5)-1-amino-3-*O*-benzoyl-4-benzyloxycarbonylamino-1-*N*,6-*O*-carbonyl-3,5,6-cyclohexanetriol (**11**) in 60% yield, together with *trans*-cyclic carbamate (**12**) in 9% yield. The structure of **11** was confirmed as follows; i) the compound (**13**), obtained by removal of the benzoyl group in **11** followed by acetylation, showed one equatorial acetyl group (δ 1.96) and one axial acetyl group (δ 2.11) in ^{1}H NMR ($CDCl_3$), ii) the compound (**14**), obtained by removal of both the benzoyl group and the cyclic carbamate in **11** followed by protection of the amino function with benzyloxycarbonyl group and finally acetylation of the hydroxyl functions, showed one equatorial acetyl group (δ 1.90) and two axial acetyl groups (δ 2.05) in ^{1}H NMR ($CDCl_3$)[13]. Thus, we achieved the inversion of the equatorial hydroxyl group in **6** described above.

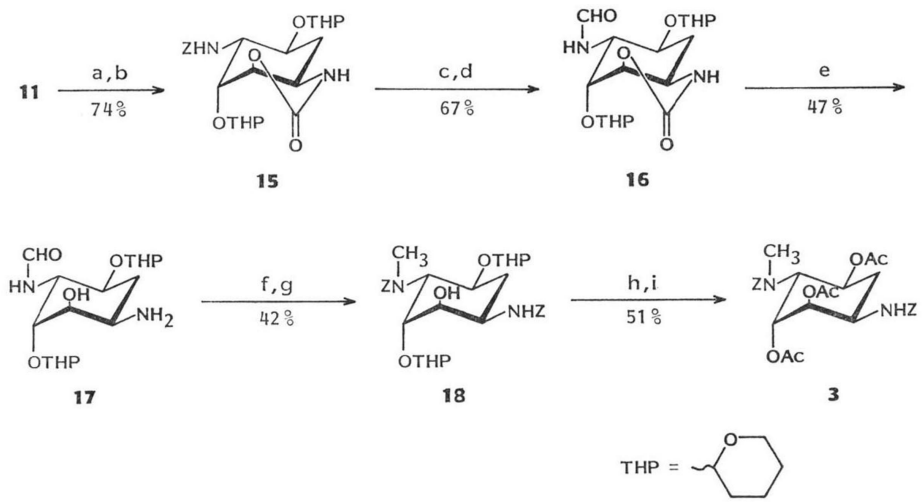
Scheme 3. Inversion step.



a: Tf_2O - pyridine, b: Δ - DMF, c: NH_4OH - MeOH, d: Ac_2O - pyridine, e: $Ba(OH)_2 \cdot 8H_2O$ - aq dioxane, f: ZCl - base.

* The configuration at the 2-position of **9** was not determined.

Scheme 4. Methylation step.

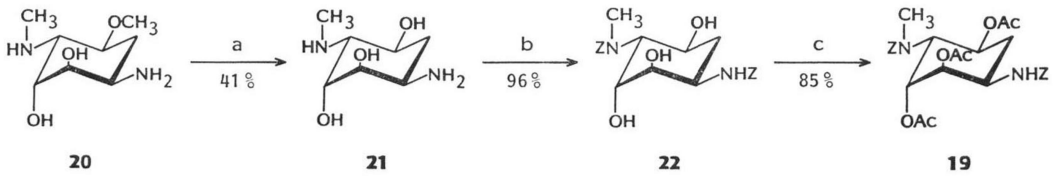


a: NH_4OH - MeOH, b: 2,3-dihydropyran-$CH_3C_6H_4SO_3H$ - DMF, c: H_2-Pd·C - aq MeOH, d: *p*-O_2N-C_6H_4OCHO-Et_3N - MeOH, e: $Ba(OH)_2 \cdot 8H_2O$ - aq dioxane, f: $LiAlH_4$ - THF, g: ZCl - base, h: $CH_3C_6H_4$-SO_3H·pyridine - EtOH, i: Ac_2O - pyridine.

The final step required 4-*N*-methylation. The *N*-methylation was carried out by reduction of a carbonyl group at the 4-amino function. For this purpose, selective removal of the cyclic carbamate is necessary. Removal of the benzoyl group in **11** by alkaline hydrolysis, followed by treatment with dihydropyran, gave the corresponding 3,5-bis-*O*-tetrahydropyranyl derivative (**15**) in 74% yield. Selective removal of the cyclic carbamate in **15** was attempted. Treatment of **15** with barium hydroxide in several conditions did not afford desirable selectivity. In order to discriminate between both protective groups, the benzyloxycarbonyl group in the 4-amino function in **15** was converted into the formyl group, which could be transformed into an *N*-methyl group by reduction. Thus, hydrogenation of **15**, followed by treatment with *p*-nitrophenyl formate, gave the corresponding 4-formamido derivative (**16**) in 67% yield. An alternative trial of selective removal of the cyclic carbamate in **16** with one equivalent of barium hydroxide octahydrate in 50% aqueous 1,4-dioxane at 55~60°C for 7 hours gave D,L-(1,3,6/4,5)-1-amino-4-formamido-3,5-bis-*O*-tetrahydropyranyl-3,5,6-cyclohexanetriol (**17**) in 47% yield. Treatment of **17** with lithium aluminum hydride, followed by protection of the amino groups with benzyloxycarbonyl function, gave D,L-(1,3,6/4,5)-1,4-bis(benzyloxycarbonylamino)-4-*N*-methyl-3,5-bis-*O*-tetrahydropyranyl-3,5,6-cyclohexanetriol (**18**) in 42% yield. Removal of the tetrahydropyranyl groups in **18**, followed by acetylation of the hydroxyl groups, afforded D,L-(1,3,6/4,5)-3,5,6-tri-*O*-acetyl-1,4-bis(benzyloxycarbonylamino)-4-*N*-methyl-3,5,6-cyclohexanetriol (**3**) in 51% yield. Except for optical activity, compound **3** prepared above was identical in every respect (TLC, IR, ^1H NMR, ^{13}C NMR, and MS) with the corresponding derivative (**19**) of D-(1,3,6/4,5)-1-amino-4-methylamino-3,5,6-cyclohexanetriol (**21**) derived from sporaricin B (see Scheme 5 and Experimental).

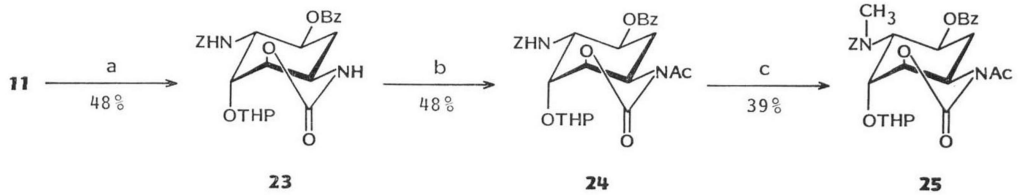
The *N*-methylation process described above, involving an exchange of the protective group at the 4-amino function in order to descriminate between both protective groups in the 1-amino group and the 4-amino group, required a lengthy sequence of reactions. To effect the necessary *N*-methylation step more conveniently, we investigated an alternative route. SANO *et al.* have reported that *N*-acetylation of D,L-(1,4/2,3,5,6)-1-amino-4-benzyloxycarbonylamino-1-*N*,2-*O*-carbonyl-5,6-*O*-isopropylidene-3-

Scheme 5. Synthesis of authentic sample.



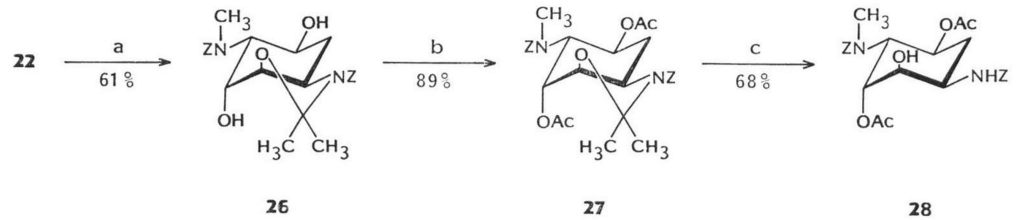
a: 56% HI, b: ZCl - base, c: Ac_2O - pyridine.

Scheme 6. Alternative methylation step.



a: 2,3-Dihydropyran-$CH_3C_6H_4SO_3H$ - DMF, b: Ac_2O - pyridine, c: MeI-Ag_2O - DMF.

Scheme 7. Transformation of aminocyclitol part for condensation reaction.



a: $(MeO)_2C(CH_3)_2$-$CH_3C_6H_4SO_3H$ - DMF, b: Ac_2O - pyridine, c: AcOH - H_2O, 4: 1.

O-tosyl-2,3,5,6-cyclohexanetetraol occurred selectively at the nitrogen atom in the cyclic carbamate[12]. Thus, we attempted to apply this method to our case. Tetrahydropyranylation of **11** gave the corresponding 5-*O*-tetrahydropyranyl ether (**23**) in 48% yield. Regioselective acetylation of **23** with acetic anhydride in pyridine afforded the expected 1-*N*-acetyl derivative (**24**) in 48% yield. Treatment of **24** with methyl iodide and silver oxide in *N*,*N*-dimethylformamide gave the corresponding 4-*N*-methyl derivative (**25**) in 39% yield. Compound **25** could be converted to **3** in three steps using standard methods. Using this alternative strategy, the *N*-methylation process was shortened from 9 steps to 6 steps and the total yield was improved slightly. Thus, synthesis of the aminocyclitol part of 3-*O*-demethylsporaricin A was completed according to the three key steps depicted in Scheme 1.

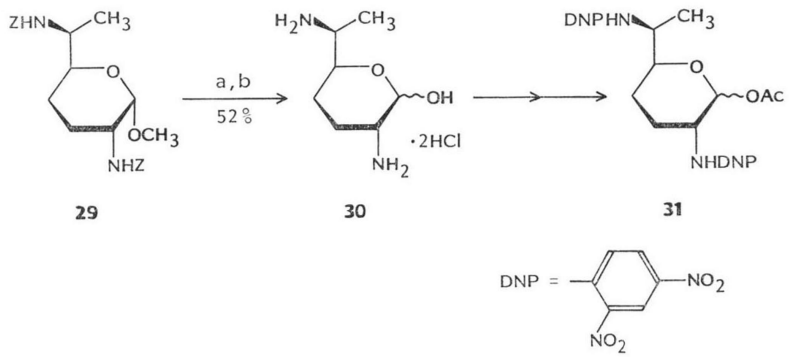
Further modification of the aglycone part is summarized in Scheme 7. Treatment of **22** with 2,2-dimethoxypropane in the presence of *p*-toluenesulfonic acid in *N*,*N*-dimethylformamide gave the corresponding 1-*N*,6-*O*-isopropylidene derivative (**26**) in 61% yield. Acetylation of **26** gave the corresponding 3,5-di-*O*-acetyl derivative (**27**) in 89% yield. Hydrolysis of **27** in 80% acetic acid afforded 1D-(1,3,6/4,5)-3,5-di-*O*-acetyl-1,4-bis(benzyloxycarbonylamino)-4-*N*-methyl-3,5,6-cyclohexanetriol (**28**) in 68% yield.

Total Synthesis of 3-*O*-Demethylsporaricin A

Synthesis of the sugar part is summarized in Scheme 8. Methyl 2,6-bis(benzyloxycarbonylamino)-2,3,4,6,7-pentadeoxy-β-L-*lyxo*-heptopyranoside (**29**)[7] was converted to 6-*epi*-purpurosamine B dihydrochloride (**30**) by hydrogenation followed by acid hydrolysis in 52% yield. Compound **30** was transformed into 1-*O*-acetyl-2,6-bis(2,4-dinitrophenylamino)-2,3,4,6,7-pentadeoxy-L-*lyxo*-heptopyranose (**31**) according to the usual method[14].

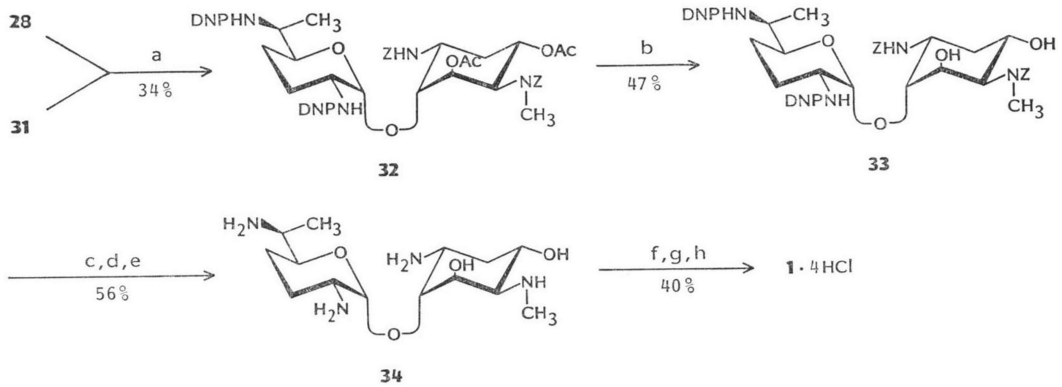
The condensation of the aglycone **28** with the sugar derivative **31** in the presence of trimethylsilyl trifluoromethanesulfonate in 1,2-dichloroethane (Scheme 9) gave the corresponding β-glycoside (**32**) in 34% yield. Removal of the acetyl groups in **32** by treatment with sodium methoxide in a mixture of 1,4-dioxane and methanol, 1: 3, gave the corresponding 3,5-diol derivative (**33**) in 47% yield. Treatment of **33** with Amberlite IRA-400 (OH^- type) resin in a mixture of acetone, water and methanol, 7: 3: 10, followed by hydrogenation and finally treatment with barium hydroxide* gave 3-*O*-demethylsporaricin B (**34**) in 56% yield. Compound **34** prepared above was identical in physical data with the

Scheme 8. Transformation of sugar part for condensation reaction.



a: H_2-Pd·C-HCl - MeOH-EtOAc, b: 2 N HCl.

Scheme 9. Synthesis of 3-*O*-demethylsporaricin A.



a: $TfOSi(CH_3)_3$-$CaSO_4$-molecular sieves 4A - $ClCH_2CH_2Cl$, b: NaOMe - MeOH, c: Amberlite IRA-400 (OH^-) - acetone-H_2O-MeOH, d: H_2-Pd·C - 1 N HCl, e: $Ba(OH)_2$ - H_2O, f: ZOSu-$Ni(OAc)_2$ - MeOH, g: $ZHNCH_2CO_2H$-$POCl_3$-DMF - aq THF - base, h: H_2-Pd·C-HCl - MeOH.

* On treatment of **33** by Amberlite IRA-400 resin, it was recognized from IR data that the cyclic carbamate was partially formed. This procedure was necessary for removal of the cyclic carbamate.

Table 1. Antibacterial activity of sporaricin analogs.

	MIC (μg/ml)[a]			
	3-*O*-Demethylsporaricin A (**1**)·4HCl		3-*O*-Demethyl-sporaricin B (**34**)	Sporaricin A
	Synthetic sample	Authentic sample		
Staphylococcus aureus 45	0.78	0.78	100	0.78
S. epidermidis 80	0.39	0.39	50	0.39
Proteus vulgaris 120	0.78	0.78	100	1.56

[a] Mueller-Hinton agar, 37°C, 18 hours; 10^6 cfu/ml.

authentic sample[4]. In biological activity, both compounds are not comparable because **34** possesses only weak antibacterial activity. Therefore, **34** was converted to 3-*O*-demethylsporaricin A (**1**) tetrahydrochloride in 40% yield according to the modified method of WATANABE *et al.*[4]. Compound **1** prepared above was identical with the authentic sample[4] derived from sporaricin B in both physical properties and biological activity as shown in Table 1.

Experimental

Melting points were measured with a Yanagimoto micro melting point apparatus and are uncorrected. Solutions were concd under reduced pressure below 50°C. Optical rotations were measured with a Jasco DIF-140 automatic polarimeter. IR spectra were recorded with a Hitachi 260-10 spectrometer. ^{1}H NMR spectra were recorded using a Jeol PMX-60, a Jeol MH-100, or a Jeol PS-100 NMR spectrometer and ^{13}C NMR spectra were recorded with a Jeol PS-100 NMR spectrometer. Chemical shifts (δ) are reported in ppm from sodium 2,2-dimethyl-2-silapentane-5-sulfonate (DSS) (in D_2O) or TMS (in $CDCl_3$, CD_3OD, or DMSO-d_6) as internal standard. EI-mass spectra were recorded with a Hitachi M-80 mass spectrometer and FD-mass spectra were recorded with a Jeol D-300 mass spectrometer.

D,L-(1,2,3/4,5,6)-3-*O*-Benzoyl-1,4-bis(benzyloxycarbonylamino)-5,6-*O*-isopropylidene-2,3,5,6-cyclohexanetetraol (**4**)

To a solution of D,L-(1,2,3/4,5,6)-1,4-bis(benzyloxycarbonylamino)-5,6-*O*-isopropylidene-2,3,5,6-cyclohexanetetraol (**2**)[11] (8.58 g) in pyridine (170 ml) was added benzoyl chloride (3.29 ml) under ice-cooling. The mixture was stirred at room temp for 21 hours. After quenched with H_2O (5 ml), the mixture was concd and dissolved in $CHCl_3$. The solution was washed successively with satd aq $NaHCO_3$ and H_2O, dried over Na_2SO_4, concd, and triturated with diisopropyl ether to give **4** (8.76 g, 84%) as a solid: mp 210~211°C; IR (Nujol) 3660~3100, 1700, 1505, 1270, 1210, 1110, 1050 cm^{-1}; ^{1}H NMR ($CDCl_3$) δ 1.35 (3H, s, CH_3), 1.60 (3H, br s, CH_3), 3.30~3.60 (1H, m), 3.80~4.40 (4H, m), 4.60~5.60 (10H, m), 7.00~7.70 (13H, m), 8.20 (2H, m); FD-MS *m/z* 591 (M^+).

D,L-(1,2,3/4,5,6)-3-*O*-Benzoyl-1,4-bis(benzyloxycarbonylamino)-2-*O*-imidazolylthiocarbonyl-5,6-*O*-isopropylidene-2,3,5,6-cyclohexanetetraol (**5**)

A mixture of **4** (1 g) and 1,1′-thiocarbonyldiimidazole (1.02 g) in THF (10 ml) was refluxed overnight. The mixture was concd and chromatographed on a silica gel column using $CHCl_3$ - EtOAc, 10: 1, as an eluant to give **5** (273 mg, 24%) as a solid: IR (Nujol) 1720, 1520 cm^{-1}; ^{1}H NMR ($CDCl_3$) δ 1.40 (3H, s, CH_3), 1.63 (3H, s, CH_3), 5.10 (2H, s, CH_2Ph), 5.20 (2H, s, CH_2Ph), 6.90 (1H, s, imidazole), 7.10~8.03 (15H, m, 3×Ph), 8.20 (1H, s, imidazole).

D,L-(1,3/4,5,6)-3-*O*-Benzoyl-1,4-bis(benzyloxycarbonylamino)-3,5,6-cyclohexanetriol (**6**) (from **5**)

A mixture of **5** (250 mg), tri-*n*-butyltin hydride (0.4 ml), and a catalytic amount of 2,2′-azobisisobutyronitrile in toluene (7.5 ml) was refluxed for 5 hours. The mixture was concd and chromato-

graphed on a silica gel column using $CHCl_3$ - EtOAc, 10: 1, to give a solid (122 mg). The solid (84 mg) was heated in 80% AcOH (5 ml) at 80°C for 1.5 hours. The mixture was concd to give **6** (71 mg, 50%) as a solid: mp 182~189°C; IR (Nujol) 3500 (sh), 3330, 1720 (sh), 1700 (sh), 1680, 1525, 1280, 1235, 1170, 1110 cm^{-1}; ^1H NMR (DMSO-d_6) δ 1.15~2.30 (2H, m, H-2), 3.25~4.15 (5H, m), 4.60~4.85 (1H, br d, OH), 4.90~5.30 (5H, m, 2×CH_2Ph and OH), 7.20~8.10 (15H, m, 3×Ph); FD-MS *m/z* 534 (M^+).

D,L-(1,2,3/4,5,6)-3-*O*-Benzoyl-1,4-bis(benzyloxycarbonylamino)-5,6-*O*-isopropylidene-2-*O*-mesyl-2,3,5,6-cyclohexanetetraol (**7**)

Mesyl chloride (0.79 ml) was added to a solution of **4** (2 g) in pyridine (20 ml) and the mixture was stirred at room temp overnight. After quenched with H_2O (1 ml), the mixture was concd and extracted with $CHCl_3$. The extract was washed successively with H_2O and satd aq $NaHCO_3$, dried over $MgSO_4$, and concd. The residue was chromatographed on a silica gel column using $CHCl_3$ - EtOAc, 5: 1, as an eluant to give **7** (1.59 g, 70%) as a syrup: IR (film) 3460~3150, 1700, 1510, 1360, 1175, 1100, 1060 cm^{-1}; ^1H NMR ($CDCl_3$) δ 1.36 (3H, s, CH_3), 1.56 (3H, s, CH_3), 3.02 (3H, s, SO_2CH_3), 3.90~4.50 (4H, m), 4.98 (2H, s, CH_2Ph), 5.10 (2H, s, CH_2Ph), 5.15~5.50 (4H, m), 7.05~7.60 (13H, m), 7.98 (2H, m); EI-MS *m/z* 668 (M^+-H).

D,L-(1,3/4,5,6)-3-*O*-Benzoyl-1,4-bis(benzyloxycarbonylamino)-2-iodo-3,5,6-cyclohexanetriol (**9**)

A solution of **7** (2 g) in 80% AcOH (40 ml) was heated at 80°C for 2 hours. The solution was poured into ice-water (200 ml). The resulted precipitates were collected by filtration, washed with H_2O until the washing was pH 7, and dried to give a crystal, mp 217~219°C. The crystal was heated with sodium iodide (17.4 g) in DMF (52 ml) at 100°C for 8 hours under nitrogen atmosphere. The mixture was poured into ice-water (200 ml). The precipitates were collected by filtration, washed with H_2O, and dissolved in $CHCl_3$. The solution was washed with H_2O, dried over Na_2SO_4, and concd to give **9** (1.94 g, 98%) as a solid: IR (Nujol) 3650~3100, 1720, 1690, 1520, 1280, 1180, 1110 cm^{-1}; ^1H NMR (DMSO-d_6) δ 3.10~4.70 (7H, m), 4.80~5.80 (7H, m), 6.80~8.10 (15H, m, 3×Ph); FD-MS *m/z* 660 (M^+).

Compound **6** (from **9**)

A solution of **9** (2.38 g) and tri-*n*-butyltin hydride (2.4 ml) was refluxed in THF in the presence of 2,2′-azobisisobutyronitrile (24 mg) for 5.5 hours under nitrogen atmosphere. The mixture was concd and triturated with *n*-hexane to give **6** (1.84 g, 96%) as a solid.

D,L-(1,3/4,5,6)-3-*O*-Benzoyl-1,4-bis(benzyloxycarbonylamino)-6-*O*-trifluoromethanesulfonyl-3,5,6-cyclohexanetriol (**10**)

To a solution of **6** (1 g) in pyridine (20 ml) was added trifluoromethanesulfonic anhydride (0.38 ml) at 0°C and the mixture was stirred at 0~5°C for 3 hours. After quenched with H_2O (1 ml), the mixture was concd and dissolved in $CHCl_3$. The organic solution was washed successively with H_2O and satd aq $NaHCO_3$, dried over $MgSO_4$, and concd to give **10** (1.18 g, 95%) as a crystal: mp 138~139°C; IR (Nujol) 3300, 1710 (sh), 1685, 1505, 1400, 1260, 1240, 1200, 1170, 1130, 1050, 1015, 940 cm^{-1}; ^1H NMR (DMSO-d_6) δ 5.05 (4H, s, 2×CH_2Ph), 7.20~8.10 (15H, m, 3×Ph).

D,L-(1,3,6/4,5)-1-Amino-3-*O*-benzoyl-4-benzyloxycarbonylamino-1-*N*,6-*O*-carbonyl-3,5,6-cyclohexanetriol (**11**) and D,L-(1,3/4,5,6)-1-Amino-3-*O*-benzoyl-4-benzyloxycarbonylamino-1-*N*,6-*O*-carbonyl-3,5,6-cyclohexanetriol (**12**)

A solution of **10** (500 mg) in DMF (10 ml) was stirred at 60~65°C for 2.5 hours under nitrogen atmosphere. The mixture was poured into ice-water (50 ml) and extracted successively with $CHCl_3$ and EtOAc. The combined extract was dried over Na_2SO_4, concd, and chromatographed on a silica gel column using $CHCl_3$ - THF, 3: 1, as an eluant. Fractions containing the material moving fast on TLC in the same solvent system were combined and concd to give **11** (192 mg, 60%) as a solid: mp 77~78°C; IR (Nujol) 3600, 3100, 1740, 1710, 1520, 1280, 1180, 1110, 1100 cm^{-1}; ^1H NMR (DMSO-d_6) δ 5.04 (2H, s, CH_2Ph), 5.70 (1H, m), 7.28~8.10 (10H, m, 2×Ph); EI-MS *m/z* 426 (M^+). Frac-

tions containing the material moving slowly on TLC were combined and concd to give **12** (29 mg, 9%) as a solid: mp 101~103°C; IR (Nujol) 3600~3100, 1740 (sh), 1720, 1520, 1275, 1180, 1095, 910 cm^{-1}; ^1H NMR (DMSO-d_6) δ 5.03 (2H, s, CH_2Ph), 7.30~8.10 (10H, m, 2×Ph); EI-MS m/z 426 (M^+).

D,L-(1,3,6/4,5)-3,5-Di-*O*-acetyl-1-amino-4-benzyloxycarbonylamino-1-*N*,6-*O*-carbonyl-3,5,6-cyclohexanetriol (**13**)

A solution of **11** (150 mg) in a mixture of 28% aq ammonia (1 ml) and MeOH (6.5 ml) was stirred at room temp for 10 hours. The solution was concd and chromatographed on a silica gel column using $CHCl_3$ - MeOH, 9: 1, as an eluant to give a crystal (72 mg), mp 213~214°C. To a solution of the crystal (43 mg) in pyridine (1.2 ml) was added acetic anhydride (0.05 ml) at room temp. The mixture was heated at 60°C for 2 hours. The mixture was concd and dissolved in $CHCl_3$ (10 ml). The solution was washed successively with 1 N HCl, satd aq $NaHCO_3$, and H_2O, dried over Na_2SO_4, and concd to give **13** (55 mg, 64%) as a solid: mp 193~195°C; IR (Nujol) 3250, 1740, 1730 (sh), 1700, 1515, 1220, 1035 cm^{-1}; ^1H NMR ($CDCl_3$) δ 1.96 (3H, s, 3-$OCOCH_3$), 2.11 (3H, s, 5-$OCOCH_3$), 5.31 (2H, s, CH_2Ph), 7.33 (5H, s, Ph); EI-MS m/z 406 (M^+).

D,L-(1,3,6/4,5)-3,5,6-Tri-*O*-acetyl-1,4-bis(benzyloxycarbonylamino)-3,5,6-cyclohexanetriol (**14**)

A solution of **11** (171 mg) and barium hydroxide octahydrate (315 mg) in 60% aq 1,4-dioxane (17 ml) was heated at 60~62°C for 5 hours. The mixture was neutralized with carbon dioxide gas and the resulted precipitates were filtered off. The filtrate was concd to give a residue, which was dissolved in 75% aq acetone (12 ml). To the solution was added benzyloxycarbonyl chloride (102 mg) under ice-cooling. After stirred at room temp for 2 hours, the mixture was poured into H_2O (10 ml). The resulted precipitates were collected by filtration, washed with EtOAc, and dried to give a solid (100 mg). The solid (76 mg) was acetylated by a similar method described in the preparation of **13** to give **14** (62 mg, 37%) as a syrup: IR (film) 3200, 1740 (sh), 1710, 1520, 1220, 1210, 1080, 1025 cm^{-1}; ^1H NMR ($CDCl_3$) δ 1.90 (3H, s, 3-$OCOCH_3$), 2.05 (6H, s, 5-$OCOCH_3$ and 6-$OCOCH_3$), 5.00 (4H, s, 2×CH_2Ph), 7.25 (10H, s, 2×Ph); FD-MS m/z 556 (M^+).

D,L-(1,3,6/4,5)-1-Amino-4-benzyloxycarbonylamino-1-*N*,6-*O*-carbonyl-3,5-bis-*O*-tetrahydropyranyl-3,5,6-cyclohexanetriol (**15**)

A solution of the 3-de-*O*-benzoyl derivative (1 g) of **11**, obtained from **11** (2.08 g) by the similar method described in the preparation of **13**, 2,3-dihydropyran (1.13 ml), and pyridinium *p*-toluenesulfonate (98 mg) in DMF (20 ml) was stirred at room temp overnight and at 60~70°C for additional 4 hours. The mixture was poured into ice-water (100 ml) and extracted with $CHCl_3$. The solution was washed with H_2O, dried over Na_2SO_4, and concd to give a syrup, which was chromatographed on a silica gel column using $CHCl_3$ - MeOH, 9: 1, as an eluant to give **15** (1.13 g, 74%) as a syrup: IR (film) 3300, 1750, 1700, 1320, 1270 cm^{-1}; ^1H NMR ($CDCl_3$) δ 5.15 (2H, s, CH_2Ph), 7.30 (5H, s, Ph); FD-MS m/z 490 (M^+).

D,L-(1,3,6/4,5)-1-Amino-1-*N*,6-*O*-carbonyl-4-formamido-3,5-bis-*O*-tetrahydropyranyl-3,5,6-cyclohexanetriol (**16**)

Compound **15** (1.1 g) was hydrogenated in a mixture of MeOH (20 ml), H_2O (4 ml), and AcOH (0.04 ml) in the presence of 10% palladium on carbon (0.5 g) at room temp under 1 atmospheric pressure of hydrogen for 4 hours. The catalyst was filtered off and the filtrate was concd to give a syrup (771 mg). To a solution of the syrup (671 mg) in 50% aq MeOH (30 ml) was added Dowex 1X2 resin (OH^- type, 1 ml) and the mixture was stirred at room temp. The resin was filtered off and the filtrate was concd to give a residue. To the solution of the residue in MeOH (15 ml) was added *p*-nitrophenyl formate (685 mg) and triethylamine (0.15 ml) at room temp. After stirred for 3.5 hours at the same temp, the mixture was concd and dissolved in $CHCl_3$. The solution was washed successively with 1 N NaOH, 1 N HCl, and H_2O, dried over Na_2SO_4, concd, and chromatographed on a silica gel column using $CHCl_3$ - acetone, 2: 1, as an eluant to give **16** (412 mg, 67%) as a glass: IR (Nujol+$CHCl_3$) 3250, 1740, 1660, 1520, 1210 cm^{-1}; ^1H NMR ($CDCl_3$) δ 8.20 (1H, s, CHO); FD-MS m/z 385 ($M^+ + H$).

D,L-(1,3,6/4,5)-1-Amino-4-formamido-3,5-bis-*O*-tetrahydropyranyl-3,5,6-cyclohexanetriol (**17**)

A solution of **16** (80 mg) and barium hydroxide octahydrate (63 mg) in 50% 1,4-dioxane (4 ml) was heated at 55~60°C for 7 hours. Carbon dioxide gas was bubbled into the mixture until the mixture was neutral. The insoluble material was filtered off. The filtrate was concd, dissolved in H_2O (20 ml), passed through a column of Amberlite IRC-50 (H^+ type, 10 ml), and eluted successively with H_2O and 1 N aq ammonia to give **17** (35 mg, 47%) as a glass: IR (Nujol) 1650, 1570~1540, 1200 cm^{-1}; ^1H NMR (D_2O) δ 8.15 (1H, s, CHO); FD-MS *m/z* 358 (M^+).

D,L-(1,3,6/4,5)-1,4-Bis(benzyloxycarbonylamino)-4-*N*-methyl-3,5-bis-*O*-tetrahydropyranyl-3,5,6-cyclohexanetriol (**18**)

To a solution of **17** (293 mg) in THF (12 ml) was added lithium aluminum hydride (124 mg) and the mixture was refluxed for 2 hours. Under ice-cooling, EtOAc (2 ml) was added to the mixture. The insoluble material was filtered off. The filtrate was concd, dissolved in H_2O, passed through a column of Amberlite IRC-50 (H^+ type), and eluted successively with H_2O and 2 N aq ammonia to give a crude residue (258 mg). To a solution of the residue (258 mg) in a mixture of acetone (4 ml) and 1 N NaOH (2 ml) was added benzyloxycarbonyl chloride (0.38 ml) under ice-cooling. After stirred for 2 hours at the same temp, the mixture was concd and dissolved in $CHCl_3$. The organic solution was washed with H_2O, dried over Na_2SO_4, concd, and chromatographed on a silica gel column using $CHCl_3$ - MeOH, 50: 1, as an eluant to give **18** (211 mg, 42%) as a syrup: IR (Nujol+EtOH) 1680, 1500, 1320, 1200 cm^{-1}; ^1H NMR ($CDCl_3$) δ 2.97 (3H, s, CH_3), 7.25 (10H, s, 2×Ph); FD-MS *m/z* 613 (M^+).

D,L-(1,3,6/4,5)-3,5,6-Tri-*O*-acetyl-1,4-bis(benzyloxycarbonylamino)-4-*N*-methyl-3,5,6-cyclohexanetriol (**3**)

A solution of **18** (200 mg) and pyridinium *p*-toluenesulfonate (482 mg) in EtOH (6 ml) was heated at 55~60°C overnight. The solution was concd and chromatographed on a silica gel column using $CHCl_3$ - MeOH, 9: 1, as an eluant to give the corresponding 3,5-dihydroxyl derivative (98 mg). The compound (90 mg) was heated in a mixture of acetic anhydride (0.5 ml) and pyridine (2 ml) at 80°C for 4 hours. The mixture was concd and dissolved in $CHCl_3$. The solution was washed successively with H_2O and satd aq $NaHCO_3$, dried over Na_2SO_4, concd, and chromatographed on a silica gel column using $CHCl_3$ - EtOAc, 10: 1, as an eluant to give **3** (87 mg, 51%) as a glass: IR (Nujol) 1740~1710, 1680, 1510, 1320, 1220 cm^{-1}; ^{13}C NMR ($CDCl_3$) δ 20.9, 30.3, 32.2, 45.8, 54.1, 65.3, 65.8, 67.1, 67.4, 68.0, 70.7, 71.3, 127.7, 128.2, 128.6, 136.0, 136.4, 155.9, 156.7, 168.7, 169.3, 169.8; EI-MS *m/z* 571 (M^+).

1D-(1,3,6/4,5)-1-Amino-4-methylamino-3,5,6-cyclohexanetriol (**21**)

A solution of 1D-(1,3,6/4,5)-1-amino-3-*O*-methyl-4-methylamino-3,5,6-cyclohexanetriol (**20**)[3] (1.17 g) in 56% hydroiodic acid (20 ml) was stirred at 100°C for 2 hours. The mixture was concd and dissolved in H_2O (20 ml). The solution was adjusted at pH 6 with 2 N aq ammonia, passed through a column of Amberlite IRC-50 (NH_4^+ type, 40 ml), and eluted successively with H_2O, 0.5 N and 1 N aq ammonia to give **21** (445 mg, 41%) as a solid: mp 163~166°C; $[\alpha]_D^{20}$ +123° (*c* 1.0, H_2O); ^1H NMR (D_2O) δ 2.43 (3H, s, CH_3).

1D-(1,3,6/4,5)-1,4-Bis(benzyloxycarbonylamino)-4-*N*-methyl-3,5,6-cyclohexanetriol (**22**)

To a solution of **21** (420 mg) in a mixture of H_2O (10 ml) and THF (10 ml) was dropwise added a solution of benzyloxycarbonyl chloride (0.8 ml) in THF (3 ml) at 0~10°C with stirring, keeping the pH between 8~9 with 1 N NaOH. The solution was stirred at the same temp for 3 hours. The mixture was extracted with EtOAc. The extract was washed with satd aq NaCl, dried over $MgSO_4$, concd, and chromatographed on a silica gel column using $CHCl_3$ - MeOH, 99: 1, as an eluant to give **22** (1.01 g, 96%) as a solid: mp 128~130°C; $[\alpha]_D^{20}$ +69° (*c* 1.0, MeOH); IR (Nujol) 1670 cm^{-1}; ^1H NMR (CD_3OD) δ 3.10 (3H, s, CH_3), 5.10 (2H, s, CH_2Ph), 5.15 (2H, s, CH_2Ph), 7.33 (10H, s, 2×Ph).

1D-(1,3,6/4,5)-3,5,6-Tri-*O*-acetyl-1,4-bis(benzyloxycarbonylamino)-4-*N*-methyl-3,5,6-cyclohexanetriol (**19**)

A solution of **22** (66 mg) and acetic anhydride (0.5 ml) in pyridine (2 ml) was heated at 80°C for 4 hours. After quenched with MeOH (2 ml), the mixture was concd and chromatographed on a silica

gel column using $CHCl_3$ - MeOH, 100: 1, as an eluant to give **19** (72 mg, 85%) as a glass. Physical data (TLC, IR, ^1H NMR, ^{13}C NMR, MS) were just the same with **3**.

D,L-(1,3,6/4,5)-1-Amino-3-*O*-benzoyl-4-benzyloxycarbonylamino-1-*N*,6-*O*-carbonyl-5-*O*-tetrahydropyranyl-3,5,6-cyclohexanetriol (**23**)

A solution of **11** (100 mg), *p*-toluenesulfonic acid (4 mg), and 2,3-dihydropyran (0.07 ml) in DMF (2 ml) was stirred at room temp overnight. The mixture was poured into ice-water (10 ml) to give precipitates. The precipitates were dissolved in $CHCl_3$. The solution was washed with H_2O, dried over Na_2SO_4, concd, and chromatographed on a silica gel column using $CHCl_3$ - THF, 3: 1, as an eluant to give **23** (57 mg, 48%) as a solid: mp 142~143°C; IR (Nujol) 1760, 1740, 1710~1680, 1520~1510, 1270, 1200 cm^{-1}; ^1H NMR ($CDCl_3$) δ 5.03 (2H, s, CH_2Ph), 7.22 (5H, s, Ph); FD-MS *m/z* 511 (M^+).

D,L-(1,3,6/4,5)-1-Acetamido-3-*O*-benzoyl-4-benzyloxycarbonylamino-1-*N*,6-*O*-carbonyl-5-*O*-tetrahydropyranyl-3,5,6-cyclohexanetriol (**24**)

A solution of **23** (220 mg) in a mixture of acetic anhydride (0.24 ml) and pyridine (6.6 ml) was heated at 80°C overnight. The mixture was concd and dissolved in $CHCl_3$. The solution was washed successively with satd aq $NaHCO_3$ and H_2O, dried over Na_2SO_4, concd, and chromatographed on a silica gel column using $CHCl_3$ - EtOAc, 5: 1, to give **24** (115 mg, 48%) as a glass: IR (Nujol) 3310, 1780, 1700, 1515, 1270, 1115 cm^{-1}; ^1H NMR ($CDCl_3$) δ 2.46 (3H, s, $COCH_3$), 5.00 (2H, s, CH_2Ph); FD-MS *m/z* 554 ($M^+ + H$).

D,L-(1,3,6/4,5)-1-Acetamido-3-*O*-benzoyl-4-benzyloxycarbonylamino-1-*N*,6-*O*-carbonyl-4-*N*-methyl-5-*O*-tetrahydropyranyl-3,5,6-cyclohexanetriol (**25**)

To a solution of **24** (25 mg) and methyl iodide (1.5 ml) in DMF (1 ml) was added silver oxide (203 mg) under ice-cooling and the mixture was stirred at room temp for 2 days in the dark. The insoluble material was filtered off and washed with $CHCl_3$. The filtrate and washings were combined, concd, and chromatographed on a silica gel column using $CHCl_3$ - EtOAc, 5: 1, as an eluant to give **25** (10 mg, 39%) as a syrup: IR (Nujol+EtOH) 1750, 1700, 1650, 1445, 1270 cm^{-1}; ^1H NMR ($CDCl_3$) δ 2.46 (3H, s, $COCH_3$), 2.95 (3H, s, CH_3), 5.20 (2H, s, CH_2Ph); FD-MS *m/z* 557 (M^+).

1D-(1,3,6/4,5)-1,4-Bis(benzyloxycarbonylamino)-1-*N*,6-*O*-isopropylidene-4-*N*-methyl-3,5,6-cyclohexanetriol (**26**)

A solution of **22** (7.1 g), 2,2-dimethoxypropane (20 ml), and *p*-toluenesulfonic acid monohydrate (1 g) in DMF (80 ml) was stirred at 50~55°C for 20 hours. Triethylamine (5 ml) was added to the mixture at room temp. The mixture was concd and chromatographed on a silica gel column using $CHCl_3$ - MeOH, 99: 1, to give **26** (4.69 g, 61%) as a glass: mp 52~56°C; $[\alpha]_D^{20}$ +1.6° (*c* 1.0, MeOH); IR (Nujol) 3400~3300, 1670~1660 cm^{-1}; ^1H NMR ($CDCl_3$) δ 1.53 (3H, s, CH_3), 1.65 (3H, s, CH_3), 3.08 (3H, s, CH_3), 5.10 (2H, s, CH_2Ph), 5.17 (2H, s, CH_2Ph), 7.33 (10H, s, 2×Ph).

1D-(1,3,6/4,5)-3,5-Di-*O*-acetyl-1,4-bis(benzyloxycarbonylamino)-1-*N*,6-*O*-isopropylidene-4-*N*-methyl-3,5,6-cyclohexanetriol (**27**)

To a solution of **26** (4.53 g) in pyridine (100 ml) was dropwise added acetic anhydride (20 ml) under ice-cooling. The mixture was stirred at room temp for 60 hours. After quenched with EtOH (20 ml), the mixture was concd and chromatographed on a silica gel column using $CHCl_3$ - MeOH, 99: 1, to give **27** (4.75 g, 89%) as a solid: mp 47~49°C; $[\alpha]_D^{20}$ +54° (*c* 1.0, $CHCl_3$); IR (Nujol) 1740, 1680 cm^{-1}; ^1H NMR ($CDCl_3$) δ 1.54 (3H, s, CH_3), 1.68 (3H, s, CH_3), 1.94 (3H, s, 3-$OCOCH_3$), 2.08 (3H, s, 5-$OCOCH_3$), 2.80 (3H, s, CH_3), 5.11 (2H, s, CH_2Ph), 5.17 (2H, s, CH_2Ph), 7.34 (10H, s, 2×Ph).

1D-(1,3,6/4,5)-3,5-Di-*O*-acetyl-1,4-bis(benzyloxycarbonylamino)-4-*N*-methyl-3,5,6-cyclohexanetriol (**28**)

A solution of **27** (2.63 g) in 80% AcOH (30 ml) was heated at 80~85°C for 24 hours. The mixture was poured into H_2O (100 ml) and extracted with $CHCl_3$. The extract was washed successively with satd aq $NaHCO_3$ and H_2O, dried over $MgSO_4$, concd, and chromatographed on a silica gel column

using $CHCl_3$ - MeOH, 99: 1, as an eluant to give **28** (1.66 g, 68%) as a glass: mp 50~53°C; $[\alpha]_D^{20}$ +67° (*c* 1.0, $CHCl_3$); IR (Nujol) 3400~3300, 1740~1660 cm^{-1}; ^1H NMR ($CDCl_3$) δ 1.97 (3H, s, 3-OCO-CH_3), 2.08 (3H, s, 5-$OCOCH_3$), 2.83 (3H, s, CH_3), 5.13 (4H, s, 2×CH_2Ph), 7.40 (10H, s, 2×Ph).

2,6-Diamino-2,3,4,6,7-pentadeoxy-L-*lyxo*-heptose Dihydrochloride (6-*epi*-Purpurosamine B Dihydrochloride) (**30**)

Methyl 2,6-bis(benzyloxycarbonylamino)-2,3,4,6,7-pentadeoxy-β-L-*lyxo*-heptopyranoside (**29**)[7] (2.41 g) was hydrogenated in a mixture of MeOH (30 ml), EtOAc (30 ml), and conc HCl (6 ml) in the presence of 10% palladium on carbon (0.5 g) under 1 atmospheric pressure of hydrogen at room temp for 4 hours. The catalyst was filtered off. The filtrate was adjusted to pH 6 with 2 N aq ammonia, passed through a column of Amberlite IRC-50 (NH_4^+ type, 30 ml), and eluted successively with 0.1 N and 0.2 N aq ammonia to give a syrup (440 mg). The syrup (420 mg) was heated in 2 N HCl (20 ml) under reflux for 16 hours. The solution was concd to give crude **30** (610 mg, 52%) as a solid.

1-*O*-Acetyl-2,6-bis(2,4-dinitrophenylamino)-2,3,4,6,7-pentadeoxy-L-*lyxo*-heptopyranose (**31**)

This compound (**31**) was obtained from **30** according to the method of SUAMI and HONDA[14].

1D-(1,3,6/4,5)-3,5-Di-*O*-acetyl-1,4-bis(benzyloxycarbonylamino)-6-*O*-[(2,6-bis(2,4-dinitrophenylamino)-2,3,4,6,7-pentadeoxy-β-L-*lyxo*-heptopyranosyl)]-4-*N*-methyl-3,5,6-cyclohexanetriol (**32**)

To a mixture of **28** (0.53 g), **31** (0.54 g), drierite (4 g), and molecular sieves 4A (4 g) in 1,2-dichloroethane (20 ml) was added trimethylsilyl trifluoromethanesulfonate (0.4 ml) at room temp. The mixture was stirred at the same temp for 1 day. The insoluble material was filtered off. The filtrate was washed with satd aq $NaHCO_3$, dried over $MgSO_4$, concd, and chromatographed on a silica gel column using $CHCl_3$ as an eluant to give **32** (345 mg, 34%) as a solid: mp 82~84°C; IR (Nujol) 1740~1710 cm^{-1}; ^1H NMR ($CDCl_3$) δ 1.30 (3H, d, $J_{6',7'}$=7 Hz, H-7′), 2.82 (3H, s, CH_3).

1D-(1,3,6/4,5)-1,4-Bis(benzyloxycarbonylamino)-6-*O*-[(2,6-bis(2,4-dinitrophenylamino)-2,3,4,6,7-pentadeoxy-β-L-*lyxo*-heptopyranosyl)]-4-*N*-methyl-3,5,6-cyclohexanetriol (**33**)

To a solution of **32** (920 mg) in a mixture of 1,4-dioxane (10 ml) and MeOH (30 ml) was added 1 N NaOMe in MeOH (1 ml). The mixture was stirred at room temp for 2 hours. After quenched with AcOH (3 ml), the solution was concd and dissolved in $CHCl_3$. The solution was washed with satd aq $NaHCO_3$, dried over $MgSO_4$, concd, and chromatographed on a silica gel column using $CHCl_3$ - MeOH, 99: 1, to give **33** (395 mg, 47%) as a glass: IR (Nujol) 1710~1670 cm^{-1}; ^1H NMR ($CDCl_3$) δ 1.07 (3H, d, $J_{6',7'}$=7 Hz, H-7′), 3.42 (3H, s, CH_3).

1D-(1,3,6/4,5)-1-Amino-6-*O*-[(2,6-diamino-2,3,4,6,7-pentadeoxy-β-L-*lyxo*-heptopyranosyl)]-4-methylamino-3,5,6-cyclohexanetriol (3-*O*-Demethylsporaricin B) (**34**)

To a solution of **33** (340 mg) in a mixture of H_2O (30 ml), acetone (70 ml), and MeOH (100 ml) was added Amberlite IRA-400 (OH^- type, 100 ml). The suspension was stirred at room temp for 20 hours. The resin was filtered off and the filtrate was concd to give a syrup. The syrup was hydrogenated in 1 N HCl (10 ml) in the presence of 10% palladium on carbon (300 mg) under 1 atmospheric pressure of hydrogen at room temp for 7 hours. The catalyst was filtered off and the filtrate was concd to give a syrup. The syrup was heated in 0.3 M barium hydroxide (30 ml) at 50~55°C for 5 hours. The mixture was neutralized with carbon dioxide gas and the resulted precipitates were filtered off. The filtrate was concd to give a syrup. The syrup was dissolved in H_2O, adjusted at pH 6 with 1 N HCl, passed through CM-Sephadex C-25 (NH_4^+ type, 30 ml), and eluted with aq ammonia with gradual increase in concentration (0→1 N). The fractions containing the title compound were combined, concd to about 10 ml, and lyophilized to give **34** (60 mg, 56%) as a solid: $[\alpha]_D^{24}$ +144° (*c* 2.06, H_2O) (ref 4, $[\alpha]_D^{25}$ +140° (*c* 2, H_2O)); ^1H NMR (D_2O) δ 1.02 (3H, d, $J_{6',7'}$=6.5 Hz, H-7′), 2.37 (3H, s, CH_3), 4.91 (1H, d, $J_{1',2'}$=3.4 Hz, H-1′).

1D-(1,3,6/4,5)-1-Amino-6-*O*-[(2,6-diamino-2,3,4,6,7-pentadeoxy-β-L-*lyxo*-heptopyranosyl)]-4-*N*-glycyl-4-methylamino-3,5,6-cyclohexanetriol (3-*O*-Demethylsporaricin A) (**1**) Tetrahydrochloride

Phosphorous oxychloride (0.05 ml) was dropwise added to a mixture of DMF (0.05 ml) and

THF (0.2 ml) under ice-cooling. The solution was stirred for 30 minutes at the same temp. To the solution was added *N*-benzyloxycarbonylglycine (0.11 g) and an additional 0.3 ml of THF. The mixture was stirred for 30 minutes at the same temp to give the activated *N*-benzyloxycarbonylglycine solution. On the other hand, a solution of **34** (60 mg) and nickel acetate tetrahydrate (200 mg) in MeOH (15 ml) was stirred at room temp for 1 hour. To the solution was added *N*-(benzyloxycarbonyloxy)succinimide (170 mg), and the solution was stirred at the same temp for 5 hours. After quenched by addition of 28% aq ammonia (3 ml), the mixture was concd and dissolved in EtOAc (30 ml). The solution was washed successively with 3 N aq ammonia and H_2O, dried over $MgSO_4$, concd, and chromatographed on a silica gel column using $CHCl_3$ - MeOH, 4: 1, to give a solid (120 mg). To a solution of the solid (120 mg) in 50% THF (20 ml) was dropwise added the activated *N*-benzyloxycarbonylglycine solution at 0~10°C with stirring, keeping the pH between 8~9 with 1 N NaOH. After stirred for 2 hours at the same temp, the mixture was extracted with EtOAc. The extract was washed with H_2O, dried over $MgSO_4$, concd, and chromatographed on a silica gel column using $CHCl_3$ - MeOH, 49: 1, to give a solid. This product was hydrogenated in a mixture of conc HCl (0.5 ml) and MeOH (3 ml) in the presence of 10% palladium on carbon (50 mg) under 1 atmospheric pressure of hydrogen at room temp for 4 hours. The catalyst was filtered off and the filtrate was concd to give a syrup. The syrup was dissolved in H_2O (10 ml) and lyophilized to give **1** as tetrahydrochloride (55 mg, 40%) as a solid: mp 220°C (dec); $[\alpha]_D^{24}$ +112° (*c* 2, H_2O); ^1H NMR (D_2O) δ 1.32 (3H, d, $J_{6',7'}$=7 Hz, H-7′), 3.06 (3H, s, CH_3), 4.02 (2H, s, CH_2NH_2), 5.43 (1H, d, $J_{1',2'}$=3 Hz, H-1′); FD-MS *m/z* 376 (M^+ + H).

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