  Bull. Chem. Soc. Jpn., 60, 713—720 (1987)

Syntheses of 2′,3′-Dideoxykanamycin A, 2′,3′-Dideoxyamikacin and Related Substances

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(Received August 19, 1986)

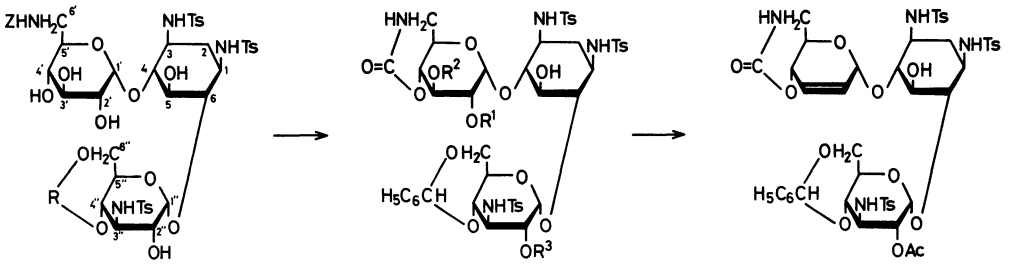
2′,3′-Dideoxykanamycin A has been prepared via two ways. 2″-*O*-Acetyl-4″,6″-*O*-benzylidene-6′-*N*,4′-*O*-carbonyl-1,3,3″-tri-*N*-tosylkanamycin A was converted to a 2′,3′-unsaturated compound by a modified Tipson and Cohen method. Hydrogenation followed by deblocking gave 2′,3′-dideoxykanamycin A (**12**). Another route for **12** was through 2′,3′-anhydro-4′,2″,4″,6″-tetra-*O*-benzoyl-2′-epi-1,3,6′,3″-tetrakis(*N*-ethoxycarbonyl)kanamycin A (**16**). Epoxide-ring opening of **16** with hydrogen iodide gave the 3′,2′-iodohydrin, which, after 2′-*O*-mesylation, led to the 2′,3′-unsaturated compound (**19**). Deblocking and hydrogenation of **19** gave **12**. 2′,3′-Dideoxykanamycin A thus synthesized was led to 2′,3′-dideoxyamikacin and other related compounds by amino protection of **12** other than the 1-amino group by zinc acetate-ethyl trifluoroacetate method followed by 1-*N*-acyl or 1-urethane formation with appropriate reagents. Their antibacterial activities were described.

Due to the lack of the hydroxyl group at C-3′, 3′-deoxykanamycin A[1] inhibits the growth of resistant bacteria producing 3′-*O*-phosphotransferases or 3′-*O*-adenylyltransferases. Attachment of (*S*)-4-amino-2-hydroxybutyryl residue at the 1-amino group of 3′-deoxykanamycin A gave 3′-deoxyamikacin,[2] which was more active than 3′-deoxykanamycin A and amikacin (1-*N*-[(*S*)-4-amino-2-hydroxybutyryl]kanamycin A). 2′-Deoxyamikacin[3] prepared from 2′-deoxykanamycin A[4] had been found more active than amikacin against a variety of resistant bacteria. This suggests the possibility that the lack of 2′-hydroxyl group of kanamycin A may deprive the hydrogen-bond formation between 5- and 2′-hydroxyl groups and cause changes of the angles (ψ and ϕ[5]) between 2-deoxystreptamine and 6-amino-6-deoxy-α-D-glucopyranose moiety to improve the antibacterial activity. The results and the assumption described above let us to prepare 2′,3′-dideoxyamikacin.

2′,3′-Dideoxykanamycin A was prepared by two different ways. 6′-*N*-Benzyloxycarbonyl-1,3,3″-tri-*N*-tosylkanamycin A[6] (**1**) was treated with benzaldehyde dimethyl acetal in an acidic medium to give the 4″,6″-*O*-benzylidene derivative (**2**) in high yield. No benzylidenation of other hydroxyl groups of **1** was observed. Treatment of **2** with sodium hydride[7] in *N*,*N*-dimethylformamide (DMF) gave the 4′,6′-cyclic carbamate (**3**), the structure being confirmed by the IR spectrum (1700 cm^{-1}[7]). Protection of the hydroxyl groups of **3** with 1,1-dimethoxycyclohexane in the presence of trifluoroacetic acid in DMF gave the 2′,3′-*O*-cyclohexylidene derivative (**4**) in high yield. In this reaction, no formation of the 5,2′-*O*-cyclohexylidene derivative as experienced in the isopropylidenation of 6′-*N*,4′-*O*-carbonyl-4″,6″-*O*-cyclohexylidene-1,3,3″-tri-*N*-tosylkanamycin A[6] was observed. Attempts to prepare 2′,3′:4″,6″-di-*O*-cyclohexylidene derivative, instead of **4**, by one-pot cyclohexylidenation of 6′-*N*,4′-*O*-carbonyl-1,3,3″-tri-*N*-tosylkanamycin A gave no desired product in pure state. In the reaction to make **4** from **3**, conversion of the benzylidene to a cyclohexylidene group was not observed, but in strongly acidic conditions some conversion was observed. After acetylation, the 2″-*O*-acetyl derivative (**5**) was hydrolyzed with 80% aqueous acetic acid (40 °C for 3.5 h) to give the decyclohexylidene derivative (**6**) in a moderate yield (57%) with the starting material (**5**) and the debenzylidene-decyclohexylidene derivative (**7**). However, removal of the both protecting groups of **5** (to give **7**) followed by rebenzylidenation gave **6** in high yield (>90%).

Double bond between C-2′ and C-3′ of **6** was created by a modified Tipson-Cohen procedure.[8] Treatment of **6** with trifluoromethanesulfonic anhydride in pyridine gave the bis(trifluoromethanesulfonate) (**8**), that was treated with sodium iodide in DMF in the absence[8] of zinc dust to give the 2′,3′-unsaturated compound **9** almost quantitatively, the yield being unexpectedly high in comparison to the yields for 2,3-unsatunation (≈50%) using other usual 2,3-bis(sulfonic ester). If zinc dust was added in the above reaction, the yield of **9** fairly decreased. In the ^1H NMR spectrum of **9**, resonances of H-2′ and H-3′ appeared at δ 5.93 and 5.80 with the $J_{2',3'}$=10 Hz (see Experimental), the shifts and the coupling being typical for 2,3-unsaturated sugars.[9] From the ^1H-shift-correlated 2-D spectrum of **9**, the sequence of H-1-H-2-H-3 was confirmed. Catalytic hydrogenation of **9** in the presence of platinum oxide gave the 2′,3′-dideoxy derivative (**10**). Alkaline treatment of **10** cleaved the both 4′,6′-carbamate and the 2″-*O*-acetyl groups to give the ninhydrin-positive derivative (**11**), which was treated with sodium in liquid ammonia to cleave the *N*-tosyl and 4″,6″-*O*-benzylidene groups to give the final product, 2′,3′-dideoxykanamycin A (**12**).

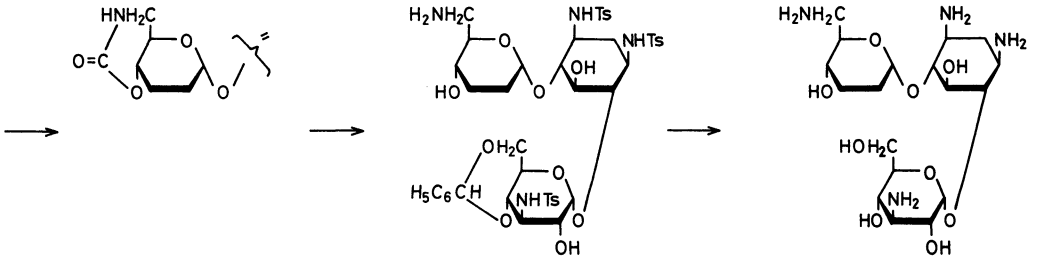
Another route for **12** was started from 3′,4′,2″,4″,6″-penta-*O*-benzoyl-1,3,6′,3″-tetrakis(*N*-ethoxycarbonyl)-kanamycin A[4] (**13**). Treatment of **13** with methanesul-



	R
1	H, H
2	CHC_6H_5

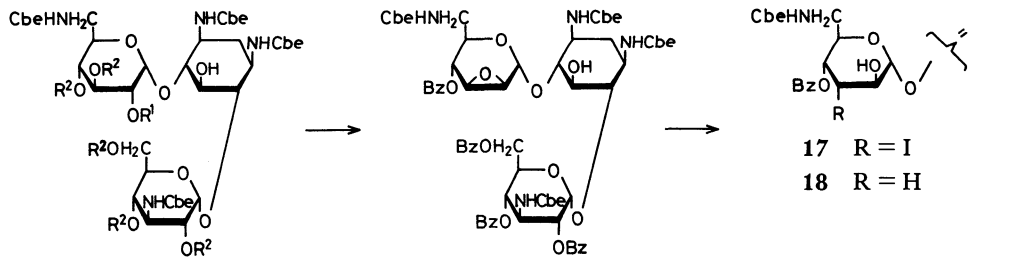
	R^1	R^2	R^3
3	H	H	H
4	$C(CH_2)_5$		H
5	$C(CH_2)_5$		Ac
6	H	H	Ac
8	SO_2CF_3	SO_2CF_3	Ac

9



10 **11** **12**

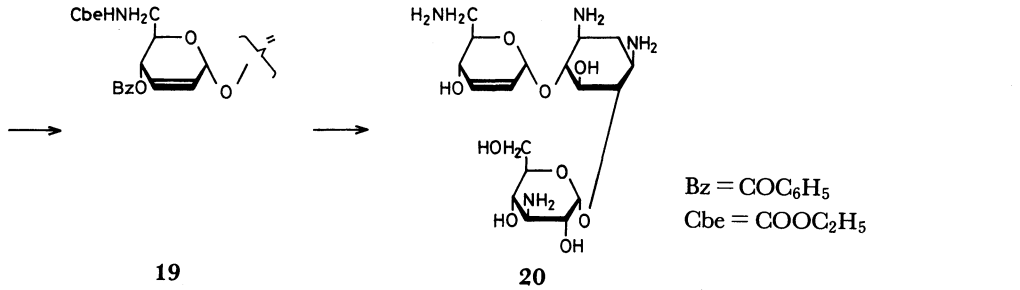
$Z = CO_2CH_2C_6H_5$



	R^1	R^2
13	H	Bz
14	SO_2CH_3	Bz
15	SO_2CH_3	H

16

17 R = I
18 R = H



19 **20**

$Bz = COC_6H_5$
$Cbe = COOC_2H_5$

fonyl chloride in pyridine gave the 2′-*O*-mesylate (**14**). Selective removal of the benzoyl groups of **14** with sodium methoxide in methanol with the mesyloxy group remained intact gave the debenzoyl-2′-*O*-mesylate (**15**). Treatment of **14** with sodium methoxide in 1,4-dioxane-methanol gave the 2′,3′-epoxide, which, without purification, was benzoylated to afford the tetrabenzoate (**16**). Epoxide-ring opening of **16** with sodium iodide in acetone gave the 3′,2′-iodohydrin (**17**). Presence of the 3′-iodo and 2′-hydroxyl (axial) groups were determined by the ^1H NMR spectrum of the reduction product **18** pro-

duced by treating **17** with tributylstannane and 2,2′-azobis(isobutyronitrile) (AIBN). The structure of **18** was further confirmed by giving, after deprotection, 3′-deoxy-2′-epikanamycin A.[10]

Mesylation of **17** with methanesulfonyl chloride in pyridine in the presence of 4-dimethylaminopyridine and triethylamine readily gave the 2′,3′-unsaturated product **19**, possively through the 2′-*O*-mesylate and subsequent 2′,3′-trans-diaxial elimination. It is interesting to note that in the epoxide-ring opening of 3′,4′-anhydro-2′,2″-di-*O*-benzoyl-4″,6″-*O*-cyclohexylidene-4′-epi-tetrakis(*N*-*t*-butoxycarbonyl)kanamycin A,[11] the 4′,3′-diequatorial-iodohydrin was produced, and the iodohydrin gave the 3′,4′-unsaturated compound by a similar procedure as described, but only at an elevated temperature (90 °C). Deblocking of **19** gave 2′,3′-dideoxy-2′-enokanamycin A (**20**). Catalytic hydrogenation of **20** gave 2′,3′-dideoxykanamycin A (**12**) identical with the specimen obtained by the previous route. Total yields of **12** starting from kanamycin A via the former and the latter routes were 17 (12 steps) and 10% (10 steps), respectively.

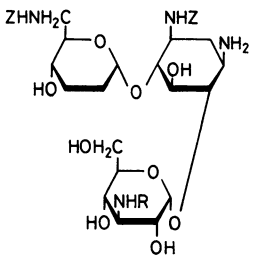
Couplings of (*S*)-4-amino-2-hydroxybutyric acid and the related acids to the 1-amino group of 2′,3′-dideoxykanamycin A were performed by utilizing zinc acetate-ethyl trifluoroacetate method.[12] Treatment of **12** with *N*-(benzyloxycarbonyloxy)succinimide[13] in dimethyl sulfoxide (DMSO) or DMF-water (10:1) in the presence of zinc acetate gave the 3,6′-bis(*N*-benzyloxycarbonyl) derivative (**21**) in 88 and 79% yields, the latter procedure being superior to the former in easy removal of the solvent. After trifluoroacetylation[12] of the 3″-amino group with ethyl trifluoroacetate in DMF (DMSO was used in the original procedure[12]), the free 1-amino group of the resulting product **22** was acylated with the active ester[13] of (*S*)-4-benzyloxycarbonylamino-2-hydroxybutyric acid. Removal of the *N*-trifluoroacetyl group of the coupling product with aqueous ammonia, followed by removal of benzyloxycarbonyl groups by catalytic hydrogenolysis gave the final product 2′,3′-dideoxyamikacin (**23**). Similarly, 1-*N*-[(*S*)-3-amino-2-hydroxypropionyl]- (**24**) and 1-*N*-[(*S*)-4-amino-2-methoxybutyryl]-2′,3′-dideoxykanamycin A (**25**) were prepared, the latter being prepared to check the biological role of the free 2‴-hydroxyl group of **23**. (*S*)-4-Benzyloxycarbonylamino-2-methoxybutyric acid (**28**) required for the synthesis of **25** was prepared from (*S*)-4-benzyloxycarbonylamino-2-hydroxybutyric acid[13] (**26**) by methylation (to give **27**) and hydrolysis.

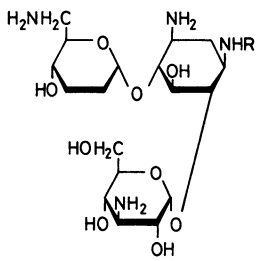
Since it was reported[14,15] that 1-*N*-(2-aminoethoxycarbonyl)kanamycin A and 1-*N*-(3-aminopropoxycarbonyl)kanamycin A, both prepared readily, showed significant antibacterial activities, 2′,3′-dideoxykanamycin A derivatives having the same 1-*N*-residues were also prepared. Treatment of **22** with *N*-[2-(benzyloxycarbonylamino)ethoxycarbonyloxy]succinimide[14] or *N*-[3-(benzyloxycarbonylamino)propoxycarbonyloxy]succinimde[14] gave the corresponding 1-*N*-alkoxycarbonyl derivatives, which, after deblocking, were led to the 1-*N*-(2-aminoethoxycarbonyl)- (**29**) and 1-*N*-(3-aminopropoxycarbonyl)-2′,3′-dideoxykanamycin A (**30**).

Antibacterial activities of the final products prepared are shown in Table 1. Both 1-*N*-[(*S*)-4-amino-2-hydroxybutyryl]- (**23**) and 1-*N*-[(*S*)-3-amino-2-hydroxypropionyl]-2′,3′-dideoxykanamycin A (**24**) showed slightly enhanced antibacterial activities in comparison to the activity of amikacin, and the two analogues (**29** and **30**) having the urethane residue at C-1 showed slightly decreased activities. The derivative (**25**) having the 2‴-*O*-methyl substituent, however, showed very weak antibacterial activity suggesting the importance of the 2‴-hydroxyl group in terms of antibacterial activity.

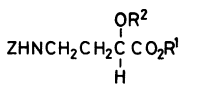
Experimental

Melting points were determined with a Kofler block and are uncorrected. Optical rotations were measured with a Perkin-Elmer 241 polarimeter. TLC was carried out on Wakogel B-5 silica gel with detection by spraying with sulfuric acid, followed by slight heating. Column chromatography was performed on Wakogel C-200. IR Spectra were measured with a JASCO A-202 grating spectrophotometer. ^{1}H NMR Spectra were recorded at 250 (unless otherwise





R



	R^1	R^2
26	H	H
27	CH_3	CH_3
28	H	CH_3

21 R = H
22 R = $COCF_3$

23 (*S*)$COCH(OH)CH_2CH_2NH_2$
24 (*S*)$COCH(OH)CH_2NH_2$
25 (*S*)$COCH(OCH_3)CH_2CH_2NH_2$
29 $COOCH_2CH_2NH_2$
30 $COOCH_2CH_2CH_2NH_2$

Table 1. Minimal Inhibitory Concentration ($\mu g\,ml^{-1}$) of **12**, **23**, **24**, **29**, and **30** with Kanamycin A (KMA) and Amikacin (AMK)

		KMA	12	AMK	23	24	29	30
S.a.	FDA 209P	1.56	1.56	0.78	0.39	0.78	1.56	0.78
S.a.	Smith	0.39	0.39	0.39	<0.2	<0.2	0.78	0.78
S.a.	Ap01 (C)	25	3.12	3.12	0.78	1.56	3.12	1.56
B.s.	PCI 219	0.78	0.39	0.39	<0.2	0.2	0.39	0.78
E.c.	K-12	0.78	0.78	0.2	0.39	0.39	1.56	0.78
E.c.	K-12 ML1629 (A)	>100	1.56	0.78	0.78	0.78	3.12	3.12
E.c.	K-12 LA290 R55 (D)	50	25	3.12	0.78	0.78	3.12	3.12
E.c.	W677	1.56	0.78	1.56	0.78	1.56	1.56	1.56
E.c.	JR66/W677 (B, D)	>100	50	1.56	3.12	3.12	6.25	3.12
E.c.	JR225 (E)	3.12	3.12	0.39	0.39	0.78	1.56	0.78
M.s.	ATCC 607	6.25	1.56	0.78	0.78	0.78	1.56	1.56
K.p.	PCI 602	1.56	3.12	1.56	1.56	1.56	3.12	3.12
K.p.	22 #3038 (B, D)	>100	50	6.25	3.12	3.12	6.25	6.25
P.v.	OX19	0.78	0.78	0.39	0.78	0.78	1.56	0.78
P.r.	GN311	0.78	0.78	0.78	1.56	1.56	1.56	0.39
S.m.		12.5	6.25	6.25	6.25	6.25	6.25	6.25
Pr. sp.	Pv16 (F)	3.12	3.12	1.56	1.56	3.12	1.56	1.56
P.a.	A3	6.25	1.56	<0.2	0.78	0.78	1.56	1.56
P.a.	TI13 (A)	50	25	1.56	3.12	3.12	12.5	25
P.a.	GN315 (G)	>100	>100	25	25	50	>100	>100

Abbreviations of bacteria: S.a. *Staphylococcus aureus*, B.s. *Bacillus subtilis*, E.c. *Esherichia coli*, M.s. *Mycobacterium smegmatis*, K.p. *Klebsiella pneumoniae*, P.v. *Proteus vulgaris*, P.r. *Proteus rettgeri* S.m. *Serratia marcescens*, Pr. *Providencia*, P.a. *Pseudomonas aeruginosa*. The letters in parenthesis show the resistant bacteria producing 3′-phosphotransferase-I (A), 3′-phosphotransferase-II (B), 4′-adenylyltransferase (C), 2″-adenylyltransferase (D), 3-acetyltransferase (E), 2′-acetyltransferase (F), and 6′-acetyltransferase (G).

stated), 200, and 90 MHz with a Bruker WM 250, JEOL FX200, and Varian EM-390 spectrometers, respectively. The chemical shifts (δ by ppm) of the spectra were measured downfield from internal TMS.

4″,6″-*O*-Benzylidene-6′-*N*-benzyloxycarbonyl-1,3,3″-tri-*N*-tosylkanamycin A (2). A solution of the mixture of **1**[1] (1.58 g), benzaldehyde dimethyl acetal (0.27 ml) and *p*-toluenesulfonic acid (55 mg of monohydrate was dried in vacuo at 100 °C for 1 h) in DMF (8 ml) was kept at room temperature. After 7 h, another acetal (0.13 ml) was added and the solution was kept for further 30 h. The solution was poured into aqueous sodium hydrogencarbonate (saturated, 320 ml) and the precipitates were thoroughly washed with water and ether to give, after drying, a solid of **2**, 1.60 g (94%), $[\alpha]_D^{23}$ −22° (*c* 0.5, oxolane).

Found: C, 55.30; H, 5.49; N, 4.84; S, 8.28%. Calcd for $C_{54}H_{64}N_4O_{19}S_3$: C, 55.47; H, 5.52; N, 4.79; S, 8.23%.

4″,6″-*O*-Benzylidene-6′-*N*,4′-*O*-carbonyl-1,3,3″-tri-*N*-tosylkanamycin A (3). To an ice-cold solution of **2** (4.65 g) in dry DMF (93 ml) was added 50% (in oil) sodium hydride (1.72 g, 9 mol equiv. for **2** as net NaH), and the mixture was vigorously stirred in the cold for 1 h under an atmosphere of nitrogen, and then overnight at room temperature. TLC (chloroform–ethanol=6 : 1) of the solution showed a single spot (R_f 0.15; cf **2**: 0.5). Addition of 25% acetic acid (15 ml) and evaporation in vacuo, with occasional additions of toluene, gave a residue, that was washed with ether, then thoroughly with water, and dried to give a solid of **3**, 3.97 g (94%), $[\alpha]_D^{22}$ −37° (*c* 1, acetone); IR (KBr): 1700 cm^{-1} (carbamate); ^1H NMR (pyridine-d_5) δ=2.09, 2.27, and 2.35 (each 3H, s, TsX3), 5.48 (1H, s, PhCH), 5.52 (1H, d, *J*=3.5 Hz, H-1′or H-1″), 5.59 (1H, d, *J*=3.5 Hz, H-1″ or H-1′).

Found: C, 52.85; H, 5.39; N, 5.26; S, 8.81%. Calcd for $C_{47}H_{56}N_4O_{18}S_3 \cdot 0.5H_2O$: C, 52.75; H, 5.37; N, 5.24; S, 8.99%.

4″,6″-*O*-Benzylidene-6′-*N*,4′-*O*-carbonyl-2′,3′-*O*-cyclohexylidene-1,3,3″-tri-*N*-tosylkanamycin A (4). A reaction flask containing **3** (3.97 g), 1,1-dimethoxycyclohexane (10.8 ml), trifluoroacetic acid (2.1 ml), dry DMF (20 ml), and dichloromethane (400 ml) was connected to a Soxlhet type of apparatus[2] fitted with a reflux condenser, the Soxhlet extractor being filled with molecular sieves 5A (120 ml, activated at 350 °C under a stream of nitrogen), and the mixture was refluxed for 6 h. Methanol liberated during the reaction was effectively removed by the sieves. TLC (chloroform–ethanol=5 : 1) of the solution showed a single spot (R_f 0.4; cf **3**: 0.18). The reaction mixture was concentrated to a small volume (≈20 ml), and poured into an ice-cold aqueous sodium hydrogencarbonate solution (saturated). The resulting precipitates were filtered, washed with water and ether alternately several times, to give, after drying, a solid of **4**, 4.15 g (97%), $[\alpha]_D^{23}$ −33° (*c* 0.6, acetone); IR (KBr): 1700 cm^{-1}; ^1H NMR (pyridine-d_5 at 90 MHz) δ=2.12, 2.24, and 2.35 (each 3H, s, TsX3).

Found: C, 55.47; H, 5.77; N, 4.88; S, 8.30%. Calcd for $C_{53}H_{64}N_4O_{18}S_3$: C, 55.78; H, 5.65; N, 4.91; S, 8.43%.

2″-*O*-Acetyl-4″,6″-*O*-benzylidene-6′-*N*,4′-*O*-carbonyl-2′,3′-*O*-cyclohexylidene-1,3,3″-tri-*N*-tosylkanamycin A (5). To an ice-cold solution of **4** (5.26 g) in dry pyridine (105 ml) was added acetic anhydride (4.8 ml) and the solution was kept at room temperature overnight. TLC (chloroform–ethanol= 10 : 1) of the solution showed a main (**5**, R_f 0.25; cf **4**: 0.18) and trace spots (R_f 0.34 and 0.38). The reaction mixture was poured, with vigorous stirring, into an ice-cold aqueous sodium hydrogencarbonate solution (saturated, 1 l). After standing for 1 h, the resulting precipitates were filtered and dried (5.52 g). Column chromatographty of the products with chloroform–acetone (10 : 7→10 : 9, gradually changed) gave a glass, 3.15 g (64%), $[\alpha]_D^{22}$ +40° (*c* 1, acetone); IR (KBr):

1730 cm^{-1}; ^1H NMR (pyridine-d_5) δ=2.16, 2.22, and 2.34 (each 3H, s, Ts ×3), 2.52 (3H, s, Ac).

Found: C, 55.47; H, 5.75; N, 4.75; S, 7.92%. Cacld for $C_{55}H_{66}N_4O_{19}S_3 \cdot 0.5H_2O$: C, 55.40; H, 5.66; N, 4.70; S, 8.07%.

2″-*O*-Acetyl-4″,6″-*O*-benzylidene-6′-*N*,4′-*O*-carbonyl-1,3,3″-tri-*N*-tosylkanamycin A (6). A suspension of **5** (1.88 g) in 80% aqueous acetic acid (38 ml) was vigorously stirred at 40 °C for 3.5 h. TLC (chloroform-acetone=1 : 2) of the resulting solution showed spots of R_f 0.44 (**5**, slight), 0.12 (**6**, major), and 0.02 (**7**). The solution was gradually poured into an ice-cold aqueous sodium hydrogencarbonate solution (saturated, 1 l), and the precipitates were filtered, washed with water, and dried (1.44 g). Column chromatography of the products with chroloform-acetone (1 : 1→1 : 2→ acetone, gradually changed) gave a thick syrup of **6**, 0.99 g (57%), recovered **5**, 0.22 g (12%), and a solid of **7**, 0.37 g. Benzylidenation of **7** (0.37 g) in the same manner as described later (see the preparation of **6** from **7**; $C_6H_5CH(OMe)_2$ 66 μl, TosOH 14 mg, and DMF 1.9 ml, room temperature, 40 h) gave another crop of **6**, 0.35 g (totally 77%), $[\alpha]_D^{22}$ −5° (*c* 1, acetone); IR (KBr): 1700 cm^{-1}; ^1H NMR (pyridine-d_5): δ=2.14, 2.26, and 2.30 (each 3H, s, Ts×3), 2.46 (3H, s, Ac), 5.50 (1H, d, *J*=3.5 H, H-1′), 5.55 (1H, s, PhCH), 5.60 (1H, dd, *J*=3.5 and 10 Hz, H-2″), 5.97 (1H, d, *J*=3.5 Hz, H-1″). Irradiation of H-1″ collapsed the doublets of H-2″ to a doublet.

Found: C, 53.34; H, 5.21; N, 5.13; S, 8.66%. Calcd for $C_{49}H_{58}N_4O_{19}S_3$: C, 53.35; H, 5.30; N, 5.08; S, 8.72%.

2″-*O*-Acetyl-6′-*N*,4′-*O*-carbonyl-1,3,3″-tri-*N*-tosylkanamycin A (7). A suspension of **5** (1.70 g) in 80% aqueous acetic acid (34 ml) was vigorously stirred at 80 °C, whereupon clear solution was obtained after ≈5 min. After 1 h (at the same temperature), the solution was concentrated. The residue suspended in toluene was evaporated, and the residual solid was dried (60 °C, 36 h in vacuo) to give **7**, 1.48 g (quant.), $[\alpha]_D^{23}$ 0° (*c* 1, acetone); IR (KBr): 1690 cm^{-1}; ^1H NMR (pyridine-d_5 at 90 MHz) δ=2.16, 2.28, and 2.32 (each 3H, s, Ts×3), 2.51 (3H, s, Ac).

Found: C, 48.81; H, 5.41; N, 5.34%. Calcd for $C_{42}H_{54}N_4O_{19}S_3 \cdot H_2O$: C, 48.83; H, 5.46; N, 5.42%.

Benzylidenation of 7 to Give 6. Compound **7** (2.53 g) was treated with benzaldehyde dimethyl acetal (0.76 ml) in DMF (12.6 ml) in the presence of *p*-toluenesulfonic acid (190 mg) similarly as described for **2** to give a solid of **6**, 2.51 g (91%).

2″-*O*-Acetyl-4″,6″,-*O*-benzylidene-6′-*N*,4′-*O*-carbonyl-1,3,3″-tri-*N*-tosyl-2′,3′-bis(*O*-trifluoromethylsulfonyl)kanamycin A (8). To an ice-cold solution of **6** (887 mg) in dry pyridine (17.7 ml) was added trifluoromethanesulfonic anhydride (1.13 ml) and the solution was kept at the temperature for 4.5 h. TLC (chloroform-acetone=1 : 1) of the solution showed a single spot (R_f 0.25; cf **6**: 0.03). The red solution was poured into excess ice-cold aqueous sodium hydrogencarbonate and the precipitates were filtered, washed thoroughly with water, and dried (in vacuo at room temperature) to give a pale yellow solid of **8**, 1.01 g (quant.); analytical sample was purified by column chromatography with chloroform-acetone (1 : 1), $[\alpha]_D^{23}$ −32° (*c* 1, acetone); IR (KBr): 1730, 1420 cm^{-1}; ^1H NMR (pyridine-d_5) δ=2.15, 2.32, and 2.38 (each 3H, s, Ts ×3), 2.47 (3H, s, Ac).

Found: C, 44.94; H, 4.32; N, 4.21; S, 11.42%. Calcd for $C_{51}H_{56}N_4O_{23}S_5F_6$: C, 44.80; H, 4.13; N, 4.10; S, 11.72%.

2″-*O*-Acetyl-4″,6″-*O*-benzylidene-6′-*N*,4′-*O*-carbonyl-2′,3′-dideoxy-2′-eno-1,3,3″-tri-*N*-tosylkanamycin A (9). A mixture of **8** (60 mg), sodium iodide (600 mg) and imidazole (21 mg) in dry DMF (dried over molecular sieves 3A, 1.2 ml) was stirred at 100 °C for 45 min. TLC (chloroform-acetone=1 : 2) of the resulting deep red solution showed a single spot (**9**, R_f 0.58; cf **8**: 0.7). To the solution, while hot (when cooled, all contents solidified hard), was added hot chloroform (20 ml), and, after shaking for a while, the organic solution isolated was washed successively with water (10 ml×3), 0.1M (1 M= mol dm^{-3}) aqueous sodium thiosulfate (10 ml×2; to remove iodine liberated), and water (10 ml×3), dried (Na_2SO_4) and concentrated to give a glass, 46 mg (98%), $[\alpha]_D^{22}$ −12.5° (*c* 1, acetone); IR (KBr): 1710 cm^{-1}; ^1H NMR (pyridine-d_5 at 70 °C) δ=2.16, 2.25, and 2.30 (each 3H, s, Ts×3), 2.36 (3H, s, Ac), 5.73 (1H, br s, H-1′), 5.80 (1H, dt, *J*=2.5, 2.5, and 10 Hz, H-3′), 5.93 (1H, d, *J*=10 Hz, H-2′).

Found: C, 54.99; H, 5.56; N, 5.15%. Calcd for $C_{49}H_{56}N_4O_{17}S_3$: C, 55.04; H, 5.28; N, 5.24%.

2″-*O*-Acetyl-4″,6″-*O*-benzylidene-6′-*N*,4′-*O*-carbonyl-2′,3′-dideoxy-1,3,3″-tri-*N*-tosylkanamycin A (10). Platinum oxide (PtO_2, 21 mg) in oxolane-water (2 : 1, 2 ml) was activated with hydrogen (3 kg cm^{-2}, 30 min). Compound **9** (100 mg) was added and the mixture was shaken at room temperature under the same hydrogen pressure for 45 min. TLC (chloroform-acetone=1 : 1) of the solution showed a single spot (**10**, R_f 0.24; cf **9**: 0.28). The mixture was filtered and evaporated to give a glass, 97.5 mg (98%), $[\alpha]_D^{23}$ +10° (*c* 1, acetone); IR (KBr); 1700 cm^{-1}; ^1H NMR (pyridine-d_5): δ=1.6—1.85 (4H), 2.04 (1H, m), and 2.50 (1H, m)(H-2_{ax}, 2_{eq}, $2'_{ax}$, $2'_{eq}$, $3'_{ax}$, $3'_{eq}$); 2.17, 2.26, and 2.34 (each 3H, s, Ts×3), 2.51 (3H, s, Ac), 5.57 (1H, s, PhCH), 5.67 (1H, dd, *J*=3.5 and 10 Hz, H-2″), 5.73 (1H, slightly unresolved s, H-1′), 6.28 (1H, d, *J*=3.5 Hz, H-1″).

Found: C, 54.58; H, 5.54; N, 5.02; S, 8.70%. Calcd for $C_{49}H_{58}N_4O_{17}S_3$: C, 54.94; H, 5.46; N, 5.23; S, 8.98%.

2′,3′-Dideoxykanamycin A (12). From 10. To a solution of **10** (178 mg) in oxolane (2.4 ml) was added 3M aqueous sodium hydroxide (1.2 ml), and the mixture was vigorously stirred at 50 °C for 2.5 h. Concentration of the two-phase mixture gave a residue, the methanol solution of which was mixed with Dowex 50W×2 resin (NH_4^+ form, 10 ml, pretreated with methanol-water=2 : 1), and the whole mixture was poured into a column containing the same resin (1 ml). Development of the column with 3M aqueous ammonia-methanol (1 : 2) gave a ninhydrin-positive deacyl product (**11**, 167 mg). The product was purified by passing the solution in acetone through a column of Sephadex LH-20 (16 ml), that was pretreated with acetone to swell and washed with 0.5% hydrochloric acid in acetone and then with acetone. To a solution of the purified product (**11**) in liquid ammonia (≈17 ml) at −50 °C was added sodium metal (≈280 mg), and the deep-blue solution was kept for 1 h at the temperature. Addition of oxolane-water (99 : 1) until the solution became colorless, followed by gradual warming to room temperature, and evaporation under diminished pressure gave a glassy residue. To an aqueous solution of the residue was added Dowex 50W×2 resin (NH_4^+ form, 50 ml), and the mixture, after shaking for a while, was poured into a column containing the same resin (10 ml). The column was washed with water, eluted with 1M aqueous ammonia, and the eluate evaporated to give a residue (63.4 mg). The residue was chromatographed on a column of CM-Sephadex C-25 (NH_4^+ form, 32 ml) with, after washing with water, aqueous ammonia (0.01→0.15M, gradually changed) to give

a solid of **12**, 43.3 mg (51% monocarbonate), $[\alpha]_D^{23}$ +114° (*c* 1, water); $R_{\text{kanamycin A}}$ 2.0 (PPC with 1-butanol-pyridine-water-acetic acid=6:4:3:1, descending), and $R_{\text{kanamycin A}}$ 1.9 (TLC with 1-butanol-ethanol-chloroform-17% aqueous ammonia=4:7:2:7); ^{1}H NMR (20% ND_3 in D_2O) δ=1.23 (1H, q, *J*=12.5 Hz, H-2_{ax}), 1.6—2.05 (5H, H-2_{eq}, -$2'_{ax}$, -$2'_{eq}$, -$3'_{ax}$, -$3'_{eq}$), 5.02 (1H, d, *J*=3.5 Hz, H-1''), 5.31 (1H, slightly br s, H-1').

Found: C, 44.08; H, 7.33; N, 10.72%. Calcd for $C_{18}H_{36}N_4O_9 \cdot H_2CO_3$: C, 44.35; H, 7.44; N, 10.89%.

From 20. An aqueous solution (5 ml) of **20** (100 mg, monohydrate) was hydrogenated in the presence of platinum oxide (10 mg) under atmospheric pressure of hydrogen at room temperature for 3 h. After filtration, the product was purified by column chromatography of Amberlite CG-50 (NH_4^+ form, 3 ml) with 0.3M aqueous ammonia to give a solid of **12**, 91 mg (83% monocarbonate).

3',4',2'',4'',6''-Penta-*O*-benzoyl-1,3,6',3''-tetrakis(*N*-ethoxycarbonyl)-2'-*O*-methylsulfonylkanamycin A (14). To a solution of **13**[5] (3.00 g) in pyridine (30 ml) was added methanesulfonyl chloride (0.63 ml) and the solution was kept at room temperature for 1 h. After addition of water (0.2 ml) and standing for 30 min, the solution was concentrated in vacuo to dryness. The solution of the residue in ethyl acetate (100 ml) was washed with aqueous sodium hydrogencarbonate (saturated) and water, dried ($MgSO_4$), and concentrated. The residue was purified by silica-gel column chromatography (60 g) with chloroform-ethanol (50:1) to give a solid of **14**, 2.98 g (94%), $[\alpha]_D^{24}$ +102° (*c* 1, chloroform); ^{1}H NMR ($CDCl_3$ at 200 MHz) δ=2.70 (3H, s, Ms).

Found: C, 57.82; H, 5.44; N, 4.01; S, 2.39%. Calcd for $C_{66}H_{74}N_4O_{26}S$: C, 57.79; H, 5.45; N, 4.09; S, 2.33%.

1,3,6',3''-Tetrakis(*N*-ethoxycarbonyl)-2'-*O*-methylsulfonylkanamycin A (15). To a solution of **14** (137 mg) in dry methanol (5 ml) was added 28% sodium methoxide in methanol (0.1 ml) and the solution was kept at room temperature for 3 d. Resulting precipitates were filtered, washed with methanol, and dried to give a solid of **15**, 69 mg (81%), $[\alpha]_D^{20}$ +79° (*c* 1, DMF); ^{1}H NMR (pyridine-d_5-D_2O=10:1) δ=1.03 (3H), 1.12 (6H), and 1.23 (3H) (each t, $CO_2CH_2CH_3$×4); 3.42 (3H, s, SO_2CH_3), 4.83 (1H, dd, *J*=3.8 and 10 Hz, H-2'), 5.45 (1H, d, *J*=3.5 Hz, H-1''), 6.17 (1H, d, H-1'); H-1' and H-2' were confirmed by the shift-correlated 2-D spectrum.

Found: C, 42.99; H, 6.66; N, 6.17%. Calcd for $C_{31}H_{54}N_4O_{21}S \cdot H_2O$: C, 42.85; H, 6.50; N, 6.45%.

2',3'-Anhydro-4',2'',4'',6''-tetra-*O*-benzoyl-2'-epi-1,3,6',3''-tetrakis(*N*-ethoxycarbonyl)kanamycin A (16). To a solution of **14** (2.84 g) in dry 1,4-dioxane-methanol (1:1, 120 ml) was added 28% sodium methoxide in methanol (4 ml) and the solution was kept at room temperature overnight. After neutralization with 1M aqueous hydrochloric acid, the solution was concentrated to dryness. To a solution of the residue in pyridine (100 ml) was added benzoyl chloride (2.5 ml) and the solution was kept at room temperature for 1 h. Successive isolation then followed as described for **14** to give the crude product, that was purified by silica-gel column chromatography (50 g) with chloroform-ethanol (50:1) to give a solid of **16**, 1.98 g (82%), $[\alpha]_D^{20}$ +75° (*c* 1, chloroform).

Found: C, 59.66; H, 5.71; N, 4.56%. Calcd for $C_{58}H_{66}N_4O_{22}$: C, 59.47; H, 5.69; N, 4.78%.

4',2'',4'',6''-Tetra-*O*-benzoyl-3'-deoxy-2',3'-diepi-1,3,6',3''-tetrakis(*N*-ethoxycarbonyl)-3'-iodokanamycin A (17). To a solution of **16** (1.85 g) in acetone (45 ml) were added sodium iodide (1.36 g) and acetic acid (1.26 ml) and the solution was refluxed for 8 h. Concentration of the solution gave a residue, that was extracted with ethyl acetate. The solution was washed with water, dried ($MgSO_4$), and concentrated to dryness. The residue was purified by silica-gel column (50 g) with chloroform-methanol=45:1 to give a solid of **17**, 1.54 g (75%), $[\alpha]_D^{20}$ +33° (*c* 0.8, chloroform).

Found: C, 53.46; H, 5.20; N, 4.23; I, 10.01%. Calcd for $C_{58}H_{67}N_4O_{22}I$: C, 53.61; H, 5.21; N, 4.31; I, 9.78%.

4',2'',4'',6''-Tetra-*O*-benzoyl-3'-deoxy-2'-epi-1,3,6',3''-tetrakis(*N*-ethoxycarbonyl)kanamycin A (18). To a solution of **17** (300 mg) in toluene (10 ml) were added tributylstannane (0.35 ml) and AIBN (10 mg), and the mixture was heated at 70 °C for 30 min under the nitrogen atmosphere. TLC (chloroform-methanol=50:1) of the solution showed a single spot (R_f 0.07; cf **17**: 0.1). Concentration of the solution gave a syrup, that was thoroughly washed with ligroine. The syrup was purified by silica-gel column chromatography with chloroform-methanol (40:1) to give a solid of **18**, 242 mg (90%), $[\alpha]_D^{21}$ +89° (*c* 1, chloroform); ^{1}H NMR (pyridine-d_5) δ=0.75 (3H), 1.04 (6H), and 1.28 (3H) (each t, $CO_2CH_2CH_3$ ×4); 2.45 (1H, m, H-$3'_{ax}$), 2.68 (1H, m, H-$3'_{eq}$), 4.61 (1H, br, s, H-2'), 6.15 (1H, br s, H-1').

Found: C, 59.35; H, 5.96; N, 4.85%. Calcd for $C_{58}H_{68}N_4O_{22}$: C, 59.38; H, 5.84; N, 4.78%.

4',2'',4'',6''-Tetra-*O*-benzoyl-2',3'-dideoxy-2'-eno-1,3,6',3''-tetrakis(*N*-ethoxycarbonyl)kanamycin A (19). To a solution of **17** (1.00 g) in pyridine (20 ml) were added methanesulfonyl chloride (0.3 ml), 4-dimethylaminopyridine (90 mg), and triethylamine (1 ml), and the mixture was stirred at room temperature for 5 h. The mixture was poured into ice-water (50 g) and the precipitates were extracted with ethyl acetate. The organic solution was washed successively with 1M aqueous potassium hydrogensulfate, aqueous sodium hydrogencarbonate (saturated), and water, dried ($MgSO_4$), and concentrated. The residue was purified by silica-gel column chromatography with chloroform-methanol (50:1) to give a solid of **19**, 650 mg (73%), $[\alpha]_D^{22}$ +78° (*c* 0.8, chloroform).

Found: C, 60.11; H, 5.76; N, 4.80%. Calcd for $C_{58}H_{66}N_4O_{21}$: C, 60.30; H, 5.77; N, 4.85%.

2',3'-Dideoxy-2'-enokanamycin A (20). To a solution of **19** (500 mg) in methanol (10 ml) was added 28% sodium methoxide in methanol (0.3 ml) and the solution was kept at room temperature for 30 min. Concentration in vacuo gave a residue, that was dissolved in water (6 ml) containing sodium hydroxide (1.5 g). The alkaline solution was heated at 100 °C for 3 h, then neutralized with aqueous hydrochloric acid. The reaction mixture was poured into a column of Amberlite CG-50 (NH_4^+ form, 20 ml) and, after washing with water, developed with 0.3M aqueous ammonia. Ninhydrin-positive fractions were collected and concentrated to give a solid of **20**, 135 mg (67% hydrate), $[\alpha]_D^{22}$ +106° (*c* 0.7, water); ^{1}H NMR (D_2O at 200 MHz) δ=1.18 (1H, q, *J*=12.5 Hz, H-2_{ax}), 1.93 (1H, dt, *J*=4.5, 4.5, and 12.5 Hz, H-2_{eq}), 5.00 (1H, d, *J*=4 Hz, H-1''), 5.43 (1H, fairly narrow and unresolved m, H-1'), 5.85 (1H, apparently ddd, the lower triplet being intense, *J*=2.3, 2.3, and 10 Hz, H-3'), 5.98 (1H, d, the high-field signal being intense, *J*=10 Hz, H-2').

Found: C, 46.25; H, 7.77; N, 11.82%. Calcd for $C_{18}H_{34}N_4O_9 \cdot H_2O$: C, 46.14; H, 7.76; N, 11.96%.

3,6'-Bis(*N*-benzyloxycarbonyl)-2',3'-dideoxykanamycin A (21). Method A. A mixture of **12** (37.7 mg, monocarbo-

nate) and zinc acetate dihydrate (56.3 mg, 3.5 molar equiv for **12**) in dry dimethyl sulfoxide (0.4 ml) was stirred at room temperature overnight. To the resulting clear solution was added *N*-(benzyloxycarbonyloxy)succinimide[13] (40.2 mg, 2.2 molar equiv for **12**) and the solution was kept at room temperature for 40 min. Addition of excess ether gave precipitates, that was filtered and washed thoroughly with ether. A solution of the precipitates in 50% aqueous oxolane was poured into a column of Amberlite CG-50 resin (7 ml, 100—200 mesh, prewashed with 1M aqueous ammonia and water, then with 50% aqueous oxolane). The column was washed with 50% aqueous oxolane (35 ml) and developed with the same solvent mixture containing ammonia (0→1M, gradually changed). The ninhydrin-positive product **21** was eluted by the procedure; zinc ion accompanied remained in the column. The eluate was evaporated to dryness to give a solid of **21**, 47.5 mg (88% hydrate), $[\alpha]_D^{22}$ +76° (*c* 0.7, DMF), R_f 0.3 (TLC with chloroform-methanol-28% aqueous ammonia=1 : 1 : 1, lower layer was used).

Found: C, 55.48; H, 6.61; N, 7.44%. Calcd for $C_{34}H_{48}N_4O_{13}\cdot H_2O$: C, 55.26; H, 6.83; N, 7.58%.

Compound **21** obtained by the above procedure was rather unstable on account of its own basicity and on storage it was liable to change to an undesirable 1,3-ureylene derivative.[16] Therefore when stable **21** was desirable, carbon dioxide was introduced to neutralize the basicity before concentration at the final stage and **21** was isolated as the carbonate salt.

Method B. To an ice-cold solution of **12** (9.84 g, monocabonate) and zinc acetate dihydrate (24.0 g, 5.7 molar equiv for **12**) in DMF-water (10 : 1, 170 ml) was added *N*-(benzyloxycarbonyloxy)succinimide (11.88 g, 2.5 molar equiv for **12**) and the solution was kept at the temperature for 1 h, then at room temperature overnight. Concentration of the solution in vacuo gave a residue, that was chromatographed on a column of Amberlite CG-50 resin (600 ml, pretreated as described for Method A) with, after washing the column with 50% aqueous 1,4-dioxane (2.7 l), 1M ammonia in the same solvent system to give a solid of **21**, 11.20 g (79% hydrate).

3,6′-Bis(*N*-benzyloxycarbonyl)-2′,3′-dideoxy-3″-*N*-trifluoroacetylkanamycin A (22). To a solution of **21** (5.10 g, monohydrate) in DMF (70 ml) was added ethyl trifluoroacetate (2.0 ml) and the solution was kept at room temperature for 20 min. TLC (lower layer of chloroform-methanol-28% aqueous ammonia=1 : 1 : 1) of the solution showed a single spot (R_f 0.5; cf **21**: 0.3). Concentration of the solution gave a syrup, that was washed with ether to give a solid of **22**, 5.9 g.

1-*N*-[(*S*)-4-Amino-2-hydroxybutyryl]-2′,3′-dideoxykanamycin A (2′,3′-Dideoxyamikacin) (23). To a mixture of **22** (5.8 g) and anhydrous sodiom carbonate (0.6 g) in 50% aqueous oxolane (170 ml) was added *N*-[(*S*)-4-benzyloxycarbonylamino-2-hydroxybutyryloxy]succinimide[3] (3.2 g) and the resulting clear solution was kept at room temperature for 2.5 h. TLC (the same solvent system as described above for **22** was used) of the solution showed a single spot (R_f 0.63). Concentration of the solution in vacuo gave a syrup, that was thoroughly washed with water to give a solid. A solution of the solid (6.07 g) in oxolane-2.2M aqueous ammonia (1 : 1, 360 ml) was kept at room temperature overnight. TLC of the solution showed a single spot (R_f 0.43). Concentration gave a residue, that was dissolved in 50% aqueous oxolane (150 ml). After addition of palladium black (360 mg) and acetic acid (6.5 ml), the solution was hydrogenated under atmospheric pressure of hydrogen for 4 h. Filtraion and neutralization of the solution with aqueous sodium hydroxide were followed by chromatography on a column of Amberlite CG-50 resin (NH_4^+ form, 150 ml). Elution with aqueous ammonia (0→0.5M, gradually changed) gave a solid of **23**, 2.89 g (69% based on **21**, monocarbonate), $[\alpha]_D^{22}$ +87° (*c* 0.9, water); IR (KBr): 1650, 950 cm^{-1}; ^1H NMR (20% ND_3 in D_2O) δ=1.43 (1H, q, *J*=12.5 Hz, H-2_{ax}), 1.6—2.05 (7H, H-2_{eq}, -2′$_{ax}$, -2′$_{eq}$, -3′$_{ax}$, -3′$_{eq}$ and COCH(OH)CH_2CH_2-), 4.17 [1H, q, *J*=3.8 and 9.5 Hz, COC*H*(OH)], 5.07 (1H, d, *J*=3.5 Hz, H-1″), 5.30 (1H, slightly br s, H-1′).

Found: C, 44.85; H, 7.43; N, 11.38%. Calcd for $C_{22}H_{43}N_5O_{11}\cdot H_2CO_3$: C, 44.87; H, 7.37; N, 11.38%.

1-*N*-[(*S*)-3-Amino-2-hydroxypropionyl]-2′,3′-dideoxykanamycin A (24). Compound **22** (5.0 g) was treated in the same manner as described for **23** only changing the succinimide reagent to *N*-[(*S*)-3-benzyloxycarbonylamino-2-hydroxypropionyloxy]succinimide (3.6 g) (prepared in a usual manner[15] from (*S*)-3-benzyloxycarbonylamino-2-hydroxypropionic acid,[16] *N*-hydroxysuccinimide, and dicyclohexylcarbodiimide) to give a solid of **24**, 1.30 g (40% based on **21**, hydrate), $[\alpha]_D^{22}$ +97° (*c* 0.7, water); ^1H NMR (D_2O at 200 MHz) δ=1.40 (1H, q, H-2_{ax}), 1.6—2.05 (5H, H-2_{eq}, -2′$_{ax}$, -2′$_{eq}$, -3′$_{ax}$, -3′$_{eq}$), 4.15 [1H, q, *J*=5 and 8 Hz, COC*H*(OH)], 5.04 (1H, d, *J*=3.5 Hz, H-1″), 5.29 (1H, slightly br s, H-1′).

Found: C, 45.44; H, 7.61; N, 12.66%. Calcd for $C_{21}H_{41}N_5O_{11}\cdot H_2O$: C, 45.22; H, 7.79; N, 12.56%.

1-*N*-(2-Aminoethoxycarbonyl)-2′,3′-dideoxykanamycin A (29). To a solution of **22** (100 mg) and anhydrous sodium carbonate (10.5 mg) in 50% aqueous oxolane (3.3 ml) was added *N*-[2-(benzyloxycarbonylamino)ethoxycarbonyloxy]-succinimide[14] (54 mg) in oxolane (1.7 ml) and the mixture was kept at room temperature for 2 h. TLC (chloroform-methanol-28% aqueous ammonia=1 : 1 : 1, lower layer) of the solution showed a single spot (R_f 0.67; cf **22**: 0.52). Evaporation gave a residue, that was washed with water and ether, and dried to give a solid (120 mg). A solution of the solid in 1,4-dioxane-2.5M aqueous ammonia (3 : 2, 6 ml) was kept at room temperature for 30 h. The slightly turbid solution showed, on TLC, a single spot (R_f 0.53). Concentration gave a residue, that was thoroughly washed with water and dried to give a solid (75 mg). A mixture of the solid and palladium black in 1,4-dioxane-water (2 : 1, 15 ml) was hydrogenated under atmospheric pressure of hydrogen for 3 h. The crude product obtained was purified by chromatography of CM-Sephadex C-25 column (NH_4^+ form, 16 ml) with, after washing with water, aqueous ammonia (0.01→0.5M) to give a solid of **29**, 33.0 mg [43% based on **21**, bis(carbonate)], $[\alpha]_D^{21}$ +85° (*c* 1, water); IR (KBr): 1700 cm^{-1}; ^1H NMR (20% ND_3 in D_2O) δ=1.43 (1H, q, *J*=13 Hz, H-2_{ax}), 1.65—2.15 (5H, H-2_{eq}, -2′$_{ax}$, -2′$_{eq}$, -3′$_{ax}$, -3′$_{eq}$), 5.11 (1H, d, *J*=3.8 Hz, H-1″), 5.35 (1H, br s, H-1′).

Found: C, 41.95; H, 7.09; N, 10.48%. Calcd for $C_{21}H_{41}N_5O_{11}\cdot 2H_2CO_3$: C, 41.62; H, 6.84; N, 10.55%.

1-*N*-(3-Aminopropoxycarbonyl)-2′,3′-dideoxykanamycin A (30). A solution of **22** (100 mg) and anhydrous sodium carbonate (10.5 mg) in 50% aqueous oxolane (3.3 ml) was treated with *N*-[3-(benzyloxycarbonylamino)propoxycarbonyloxy]-succinimide[14] (56 mg) in oxolane (1.7 ml) in a manner as described for **29** to give a solid, which was then led to **30**, 32.0 mg [40% based on **21**, bis(carbonate)], $[\alpha]_D^{21}$ +76° (*c* 2.8, water); ^1H NMR (20% ND_3 in D_2O) δ=1.43 (1H, q, *J*=12.5 Hz, H-2_{ax}), 1.65—2.15 (7H, H-2_{eq}, -2′$_{ax}$, -2′$_{eq}$, -3′$_{ax}$, -3′$_{eq}$ and

C(O)OCH$_2$C*H*$_2$CH$_2$NH$_2$), 5.11 (1H, d, J=3.8 Hz, H-1''), 5.35 (1H, br s, H-1').

Found: C, 42.46; H, 7.08; N, 9.94%. Calcd for $C_{22}H_{43}N_5O_{11}\cdot 2H_2CO_3$: C, 42.53; H, 6.99; N, 10.34%.

(*S*)-4-Benzyloxycarbonylamino-2-methoxybutyric Acid Methyl Ester (27). To a solution of (*S*)-4-benzyloxycarbonylamino-2-hydroxybutyric acid[13] (**26**, 2.5 g) in DMF (26 ml) were added silver oxide (4.6 g) and methyl iodide (1.9 ml), and the mixture was stirred in the dark at 5 °C overnight. After addition of methanol (0.5 ml) and ethyl acetate (50 ml), the mixture was filtered, and the filtrate was evaporated. The residue was chromatographed on silica gel with benzene-ethyl acetate (3 : 1) to give an oil of **27**, 1.42 g (51%), $[\alpha]_D^{20}$ −17° (*c* .1, chloroform); ^{1}H NMR ($CDCl_3$) δ=1.75—2.1 (2H, m, CH_2-3), 3.25 (2H, t, J=6 Hz, CH_2-4), 3.35 (3H, s, OCH_3), 3.70 (3H, s, CO_2CH_3), 3.82 (1H, q, J=5 and 7 Hz, H-2), 5.05 (2H, s, $PhCH_2O$), 7.30 (5H, s, C_6H_5).

(*S*)-4-Benzyloxycarbonylamino-2-methoxybutyric Acid (28). To a solution of **27** (1.4 g) in acetone (40 ml) was added 1M aqueous sodium hydroxide (7.5 ml) and the mixture was kept at room temperature for 40 min. After neutralization to pH 9 with aqueous hydrochloric acid, the solution was concentrated. The concentrate was washed with ether and, after addition of aqueous hydrochloric acid (to pH 6), extracted with ethyl acetate. The organic solution was dried (Na_2SO_4) and concentrated to give an oil, that was purified by a charcoal column developed by methanol to give an oil of **28**, 0.85 g (64%), $[\alpha]_D^{22}$ −16° (*c* 1, chloroform); *m/z* 267.11269 (M^+), Calcd for $C_{13}H_{17}NO_5$: 267.11290; ^{1}H NMR ($CDCl_3$) δ=2.00 (2H, m, CH_2-3), 3.27 (2H, t, J=6 Hz, CH_2-4), 3.37 (3H, s, OCH_3), 3.81 (1H, t, J=6 Hz, H-2), 5.07 (2H, s, $PhCH_2O$), 7.30 (5H, s, C_6H_5).

1-*N*-[(*S*)-4-Amino-2-methoxybutyryl]-2',3'-dideoxykanamycin A (25). Compound **22** (360 mg) was treated in a similar manner as described for **23** with *N*-[(*S*)-4-benzyloxycarbonylamino-2-methoxybutyryloxy]succinimide (255 mg), prepared from **28** in a ususl manner, to give a solid of **25**, 100 mg (41% based on **21**), $[\alpha]_D^{22}$ +87° (*c* 0.7, water) and **12**, 105 mg (48%). **25**: ^{1}H NMR (D_2O at 200 MHz) δ=1.40 (1H, q, H-2$_{ax}$), 1.6—2.02 (7H, H-2$_{eq}$, -2'$_{ax}$, -2'$_{eq}$, -3'$_{ax}$, -3'$_{eq}$ and COCH(OCH$_3$)-C*H*$_2$CH$_2$-), 3.37 (3H, s, OCH_3), 5.06 (1H, d, J=3.5 Hz, H-1''), 5.28 (1H, slightly br s, H-1').

Found: C, 47.13; H, 8.00; N, 12.13%. Calcd for $C_{23}H_{45}N_5O_{11}\cdot H_2O$: C, 47.16; H, 8.10; N, 11.96%.

References

1) S. Umezawa, T. Tsuchiya, R. Muto, Y. Nishimura, and H. Umezawa, *J. Antibiot.*, **24**, 274 (1971).
2) T. Tsuchiya, T. Jikihara, T. Miyake, S. Umezawa, M. Hamada, and H. Umezawa, *J. Antibiot.*, **32**, 1351 (1979).
3) S. Fukatsu, T. Yoneta, Unpublished data.
4) N. Sakairi, T. Yoneta, M. D. Rahman, and Y. Ishido, *J. Carbohydr. Chem.*, **3**, 51 (1984).
5) P. Dais, T. K. M. Shing, and A. S. Perlin, *J. Am. Chem. Soc.*, **106**, 3082 (1984); related references are cited therein.
6) T. Jikihara, T. Tsuchiya, H. Umezawa, and S. Umezawa, *Carbohydr. Res.*, **89**, 91 (1981).
7) Y. Takagi, C. Komuro, T. Tsuchiya, and S. Umezawa, *Bull. Chem. Soc. Jpn.*, **54**, 1834 (1981); related references are cited therein.
8) T. Miyake, T. Tsuchiya, S. Umezawa, and H. Umezawa, *Carbohydr. Res.*, **49**, 141 (1976).
9) E. Albano, D. Horton, and T. Tsuchiya, *Carbohydr. Res.*, **2**, 349 (1966).
10) T. Yoneta, T. Matsuno, H. Nanahoshi, and S. Fukatsu, *Nippon Kagaku Kaishi*, **1982**, 1730.
11) T. Yoneta, T. Matsuno, and S. Fukatsu, *Chem. Pharm. Bull.*, **29**, 3464 (1981).
12) T. Tsuchiya, Y. Takagi, and S. Umezawa, *Tetrahedron Lett.*, **1979**, 4951.
13) H. Kawaguchi, T. Naito, S. Nakagawa, and K. Fujisawa, *J. Antibiot.*, **25**, 695 (1972).
14) Y. Takagi, C. Komuro, T. Tsuchiya, S. Umezawa, M. Hamada, and H. Umezawa, *J. Antibiot.*, **34**, 1 (1981).
15) A. K. Mallams, P. Reichert, and J. B. Morton, Current Chemotherapy and Infectious Disease, Proceedings of the 11th International Congress of Chemotherapy and the 19th Interscience Conference on Antimicrobial Agents and Chemotherapy, Boston, Massachusetts, U. S. A., October 1-5, 1979, Vol. 1, p. 406, 1980.
16) T. Yamaguchi, T. Tsuchiya, and S. Umezawa, *J. Antibiot.*, **30**, 71 (1977).