THE TOTAL SYNTHESIS OF KANAMYCIN C

Sir:

We wish to report the synthesis of kanamycin C[1] (VIII) from paromamine. Since we have previously synthesized paromamine[2] (I), the combined achievements constitute the first synthesis of kanamycin C.

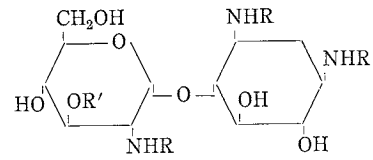
Tri-N-carbobenzoxyparomamine (II), m.p. 258°C (decomp.), $[\alpha]_D^{18}$+64.5° (c 0.67, DMF), was treated with 2,2-dimethoxypropane and *p*-toluenesulfonic acid in N,N-dimethylformamide (DMF) at 110°C to give the diisopropylidene derivative (III) in a quantitative yield; m. p. 243~245°C, $[\alpha]_D^{18}$+71° (c 0.59, DMF). Benzylation of III with benzyl bromide in the presence of barium oxide and barium hydroxide in DMF gave IV in a 82% yield; m. p. 160~162°C, $[\alpha]_D^{18}$+72° (c 0.60, DMF). Deacetonation of IV by treatment with 80% acetic acid gave quantitatively 4-O-(3-O-benzyl-2-carbobenzoxyamino-2-deoxy-α-D-glucopyranosyl)-N,N'-dicarbobenzoxy-2-deoxystreptamine (V); m.p. 270~271°C (decomp.), $[\alpha]_D^{18}$+101° (c 0.52, DMF). Partial acetonation of V with 2,2-dimethoxypropane and *p*-toluenesulfonic acid in DMF at room temperature gave monoisopropylidenemonobenzyl derivative (VI) in a 55% yield; m. p. 239~241°C, $[\alpha]_D^{18}$+75° (c 0.63, DMF).

Anal. found: C 64.29, H 6.11, N 4.78.

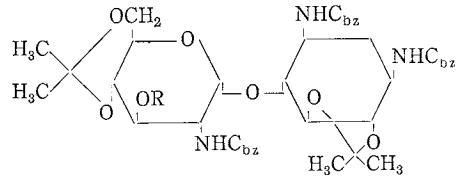
Calcd. for $C_{46}H_{53}N_3O_{13}$: C 64.55, H 6.24, N 4.91%.

Methyl 3-acetamido-3-deoxy-β-D-glucopyranoside was benzylated as described in the synthesis of IV to afford the tribenzyl derivative, which was converted to 3-acetamido-2,4,6-tri-O-benzyl-3-deoxy-α-D-glucopyranosyl chloride*, m. p. 143~144°C (decomp.), $[\alpha]_D^{23}$+78° (c 1.0, $CHCl_3$) by successive hydrolysis, acetylation and chlorination.

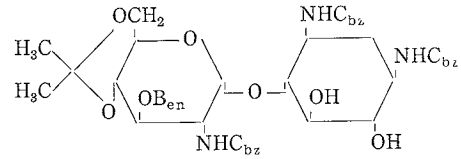
The condensation of VI with the benzylated glycosyl chloride was conducted as follows: A sample (1.12 g) of VI was



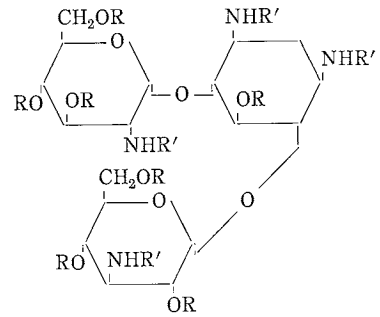
I: R, R'=H (Paromamine)
II: R=$COOCH_2C_6H_5$ R'=H
V: R=$COOCH_2C_6H_5$ R'=$CH_2C_6H_5$

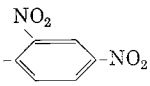


III: R=H
IV: R=$CH_2C_6H_5$



VI



VII: R=$COCH_3$ R'= 

VIII: R, R'=H (Kanamycin C)

dissolved in an anhydrous mixture (16.5 ml) of benzene-dioxane (2:1), Drierite (3.66 g) and mercuric cyanide (0.46 g); the glycosyl chloride (1.3 g) were added with stirring and the mixture was then vigorously stirred at 100°C for 6 hours to give the condensation products. The products were treated with acetic acid to remove the isopropylidene group, hydrogenated in a mixture of dioxane-water-conc. hydrochloric acid (10:2:1) over palladium black with occasional addition of water, and de-N-acetylated with barium hydroxide to give a ninhydrin-positive product. This was dinitrophenylated with 2,4-dinitrofluorobenzene in aque-

* This compound have been reported by S. UMEZAWA *et al.* at the 20th Annual Meeting of the Chemical Society of Japan, Tokyo, Mar. 31, 1967

ous ethanol in the presence of sodium bicarbonate and then O-acetylated with acetic anhydride and anhydrous sodium acetate. The resulting product, which showed about four spots with Rf-values of 0.56, 0.45, 0.35 and 0.25 on a thin-layer chromatogram (TLC) with a solvent system (A): toluene-MEK (2:1), was chromatographed on a silica-gel column (49×210 mm) with the same solvent. The substance having an Rf-value of 0.35 was isolated and recrystallized from toluene-MEK affording yellow crystals of VII; yield 285 mg (15 % over-yield from VI); m. p. 208~211°C (decomp.), $[\alpha]_D^{18}$+285° (c 0.75, acetone). IR spectrum (KBr): 3320, 1620, 1595, 1550, 1525, 1335, 835, 745 (NH-DNP), 1750, 1365, 1220 (OAc) cm^{-1}.

Anal. found: C 46.68, H 4.34, N 11.84.

Calcd. for $C_{56}H_{58}N_{12}O_{34}$: C 46.61, H 4.05, N 11.65 %.

On the other hand, kanamycin C[1] was dinitrophenylated and acetylated to give hepta-O-acetyl-tetra-N-(2,4-dinitrophenyl)-kanamycin C; m. p. 208~211°C (decomp.), $[\alpha]_D^{18}$+299° (c 0.64, acetone).

Anal. found: C 46.65, H 4.24, N 11.78.

Calcd. for $C_{56}H_{58}N_{12}O_{34}$: C 46.61, H 4.05, N 11.65 %.

On TLC with a solvent system (A), the synthetic product VII and the above-mentioned derivative of natural kanamycin C showed identical mobilities. Their infrared spectra were superimposable. Hydrolysis of VII with methanolic ammonia followed by treatment with an excess of Dowex 1×2 (OH^-) resin gave a crude free base, which was purified by chromatography on a column of Dowex 1×2 (OH^-) resin using water and recrystallized from aqueous methanol-ethanol to give a crystalline free base of VIII; $[\alpha]_D^{18}$+139° (c 0.50, water).

Anal. found: C 44.40, H 7.28, N 11.80.

Calcd. for $C_{18}H_{36}N_4O_{11}$: C 44.62, H 7.49, N 11.56 %.

The natural kanamycin C showed $[\alpha]_D^{18}$+145° (c. 0.58, water) [lit.[1], $[\alpha]_D^{20}$+126° (water)]. On descending paper chromatography by ninhydrin coloration using a solvent system: *n*-butanol - pyridine - water - acetic acid (6:4:3:1), the R-value of the synthetic product VIII agreed with that of the natural kanamycin C. Infrared spectra of VIII and the natural kanamycin C were identical. The antibiotic spectra and minimal inhibitory concentrations (MIC) of the synthetic product VIII against test organisms were in agreement with those of the natural kanamycin C as shown in Table 1.

Table 1. MIC's of synthetic (VIII) and natural kanamycin C as determined by the dilution method in bouillon, mcg/ml

Test organisms	VIII	Kanamycin C
Bacillus subtilis PCI 219	0.5	0.5
Mycobacterium tuberculosis 607	7.8	3.9
Escherichia coli	3.9	3.9
M. pyogenes var. *aureus* 209P	1.0	1.9

The details of the present work will be published in Bull. Chem. Soc. Japan.

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