Communications to the editor

SYNTHESES OF (S)-4-AMINO-2-HYDROXYBUTYRYL DERIVATIVES OF 3′,4′-DIDEOXYKANAMYCIN B AND THEIR ANTIBACTERIAL ACTIVITIES

Sir:

KAWAGUCHI *et al.*[1,2] reported that 1-N-[(S)-4-amino-2-hydroxybutyryl]-kanamycin (BB-K8) was effective against kanamycin-sensitive and -resistant bacteria, but its three positional isomers, 3-, 6′-, and 3″-N-[(S)-4-amino-2-hydroxybutyryl]-kanamycin were almost inactive. In a previous paper[3], we reported the syntheses of 1-N-[(S)-4-amino-2-hydroxybutyryl]-kanamycin B and -3′,4′-dideoxykanamycin B (1-AHB-DKB) which were active against kanamycin-resistant bacteria producing kanamycin-phosphotransferases I[4,5] and II[6], and kanamycin-nucleotidyltransferase[7~10]. In the present communication, the syntheses and characterization of four positional isomers of 1-AHB-DKB and two diacyl derivatives of 3′,4′-dideoxykanamycin B[11,12] are reported. One of the latter, 1,2′-di-N-[(S)-4-amino-2-hydroxybutyryl]-3,′4′-dideoxykanamycin B (1,2′-AHB-DKB) was active against kanamycin-sensitive and -resistant bacteria.

As described in the previous paper[3], 1-AHB-DKB was synthesized from 6′-N-*tert*-butyloxycarbonyl-3′,4′-dideoxykanamycin B (**I**) by reaction with an equimolar amount of *tert*-butyloxycarbonyl azide followed by acylation with the N-hydroxysuccinimide ester of (S)-4-*tert*-butyloxycarbonylamino-2-hydroxybutyric acid (**II**) and removal of the N-protecting group. The reaction products were adsorbed on a column of Amberlite CG 50 (NH_4^+) and separated into 1-AHB-DKB, its three positional isomers (3-, 2′-, and 3″-AHB-DKB) and two diacyl derivatives (1,2′- and 3,2′-AHB-DKB) by stepwise elution with 0.5, 0.75 and 1.0 N ammonia. After washing the column with water, unreacted DKB (32% yield) and 2′-AHB-DKB (21%) were eluted with 0.5 N ammonia, 3-AHB-DKB (6%), 1-AHB-DKB (12%) and 3″-AHB-DKB (4%) were eluted with 0.75 N ammonia, and 3,2′-AHB-DKB (3%), 1,2′-AHB-DKB (3%) and the other diacyl derivatives were eluted with 1.0 N ammonia. The 1,2′-AHB-DKB was also synthesized in good yield by reaction with **I** and 2.5 equivalents of **II**. The 3″-AHB-DKB was also prepared from 1,3,2′,6′-tetra-N-*tert*-butyloxycarbonyl-3′,4′-dideoxykanamycin B (**III**) in a 78% yield. The **III** was synthesized from DKB

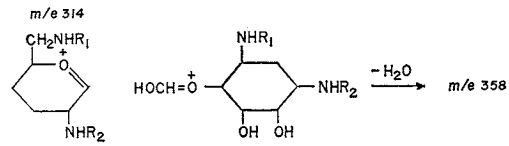
Table 1. The properties of (S)-4-amino-2-hydroxybutyryl derivatives of DKB

Derivatives	mp (dec.)	$[\alpha]_D$ in H_2O	Molecular formula*1	Rf on TLC*2	MS of N-acetyl deriv.*3 (*m/e*)	
					314	358
3-AHB-DKB	166~168°	+ 77° at 24°	$C_{22}H_{44}N_6O_{10}\cdot H_2CO_3$	0.24	−	+
2′-AHB-DKB	164~166°	+ 98° at 26°	$C_{22}H_{44}N_6O_{10}\cdot H_2CO_3$	0.29	+	−
6′-AHB-DKB	168~170°	+ 83° at 26°	$C_{22}H_{44}N_6O_{10}\cdot H_2CO_3$	0.27	+	−
3″-AHB-DKB	177~180°	+100° at 26°	$C_{22}H_{44}N_6O_{10}\cdot H_2CO_3$	0.12	−	−
1,2′-AHB-DKB	168~170°	+ 78° at 24°	$C_{26}H_{51}N_7O_{12}\cdot 2H_2CO_3$	0.09	+	+
3,2′-AHB-DKB	166~167°	+ 76° at 24°	$C_{26}H_{51}N_7O_{12}\cdot 2H_2CO_3$	0.18	+	+

*1 Satisfactory elemental analyses were obtained for all compounds.

*2 Thin-layer chromatography on Silica gel G using butanol-ethanol-chloroform-17% ammonia (4: 5: 2: 5 in volume).

*3 Penta-N-acetyl derivatives were prepared with acetic anhydride in methanol. *m/e* 314: fragment from N-[(S)-4-amino-2-hydroxybutyryl]-2, 6-diamino-2, 3, 4, 6-tetradeoxy-α-D-glucose. *m/e* 358: fragment from N-[(S)-4-amino-2-hydroxybutyryl]-2-deoxystreptamine.



R_1 = Ac, R_2 = -C(=O)-CHOH-CH_2-CH_2-NHAc

or R_1 = -C(=O)-CHOH-CH_2-CH_2-NHAc, R_2 = Ac

Table 2. The antimicrobial spectra of (S)-4-amino-2-hydroxybutyryl derivatives of DKB

Test organisms	Minimum inhibitory concentrations (mcg/ml)						
	1-AHB-DKB	3-AHB-DKB	2′-AHB-DKB	6′-AHB-DKB	3″-AHB-DKB	1,2′-AHB-DKB	3,2′-AHB-DKB
Staph. aureus FDA 209P	0.78	25	3.13	6.25	25	25	50
Staph. aureus SMITH	< 0.20				1.56	<0.20	
Staph. aureus TERAJIMA	< 0.20				<0.78	<0.20	
Sarcina lutea PCI 1001	1.56				100	1.56	
B. anthracis	< 0.20				3.13	<0.20	
B. subtilis PCI 219	< 0.20				<0.78	<0.20	
B. subtilis NRRL B-558	< 0.20				<0.78	<0.20	
B. cereus ATCC 10702	1.56				25	1.56	
Coryn. bovis 1810	0.39				25	0.78	
Mycob. smegmatis ATCC 607	< 0.20	3.13	3.13	6.25	6.25	0.20	50
Sh. dysenteriae JS 11910	6.25				>100	6.25	
Sh. flexneri 4b JS 11811	6.25				>100	6.25	
Sh. sonnei JS 11746	3.13				100	6.25	
Sal. typhosa T-63	< 0.20				25	0.78	
Sal. enteritidis 1891	1.56				50	1.56	
Prot. vulgaris OX 19	0.39				25	0.78	
Kl. pneumoniae PCI 602	0.78	25	3.13	12.5	25	0.78	100
Kl. pneumoniae 22 #3038	1.56	50	100	>100	100	1.56	>100
E. coli NIHJ	0.78	50	12.5	12.5	50	3.13	100
E. coli K-12	0.78	50	12.5	12.5	100	1.56	100
E. coli K-12 R5	0.78	25	12.5	50	50	1.56	50
E. coli K-12 ML1629	0.78	50	12.5	12.5	50	3.13	100
E. coli K-12 ML1630	0.78	50	12.5	12.5	50	3.13	100
E. coli K-12 ML1410	0.78	50	12.5	12.5	100	3.13	50
E. coli K-12 ML1410 R81	1.56	100	25	25	100	3.13	100
E. coli LA290 R55	0.78	50	100	100	100	1.56	100
E. coli LA290 R56	0.39	25	50	>100	25	0.78	100
E. coli LA290 R64	0.78	50	50	100	50	0.78	50
E. coli W677	0.39	25	6.25	6.25	25	1.56	50
E. coli JR66/W677	1.56	100	100	>100	100	6.25	>100
Ps. aeruginosa A3	3.13	>100	100	25	>100	25	>100
Ps. aeruginosa No. 12	1.56	>100	25	12.5	>100	6.25	>100
Ps. aeruginosa TI-13	3.13	>100	100	50	>100	12.5	>100
Ps. aeruginosa GN315	25	>100	>100	>100	>100	100	>100
Ps. aeruginosa 99	12.5	>100	>100	50	>100	50	>100

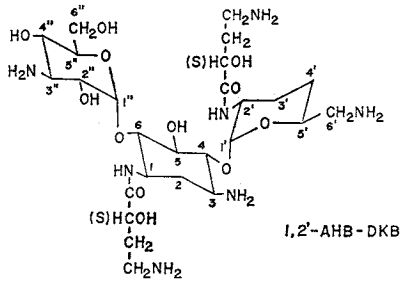
in a 50% yield by reaction with excess amounts of *tert*-butyloxycarbonyl azide in a mixture of water, pyridine and triethylamine (10: 10: 1 in volume) overnight at room temperature, followed by silicic acid chromatography, mp 210~212°C (dec), $[\alpha]_D^{26}$ +68° (*c* 2.1, dimethylformamide). Satisfactory elemental analysis for $C_{38}H_{69}N_5O_{16}$ was obtained.

The 6′-acyl derivative (6′-AHB-DKB) was synthesized from **I** in a 64% yield as follows. The tetra-N-benzyloxycarbonylation of **I** by the usual SCHOTTEN-BAUMANN procedure and debutyloxycarbonylation with 90% trifluoroacetic acid afforded 1,3,2′,3″-tetra-N-benzyloxycarbonyl-3′,4′-dideoxykanamycin B (**IV**). The **IV** without purification was acylated with the N-

hydroxysuccinimide ester of (S)-4-benzyloxycarbonyl-amino-2-hydroxybutyric acid. After removal of the protecting group by catalytic hydrogenation with 5% palladium on carbon, the 6′-AHB–DKB was purified by resin chromatography on Amberlite CG 50 (NH_4^+).

The properties of the (S)-4-amino-2-hydroxybutyryl derivatives of DKB are summarized in Table 1. The structures of these derivatives were completely confirmed by the pmr spectra, mass spectra of penta-N-acetyl derivatives (Table 1), paperchromatography of acid hydrolyzates after N-ethoxycarbonylation, and rotation of mono-N-ethoxycarbonyl-2-deoxystreptamine[3].

The antimicrobial spectra of these derivatives are shown in Table 2. The 1,2′-AHB–DKB was active against kanamycin-sensitive and -resistant bacteria, but less active than 1–AHB–DKB. Other derivatives are weakly active against bacteria. It is interesting that 1,2′-AHB–DKB is several times more active than 2′-AHB–DKB.



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