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STRUCTURES OF OA-6129A, B_1, B_2 AND C, NEW CARBAPENEM ANTIBIOTICS

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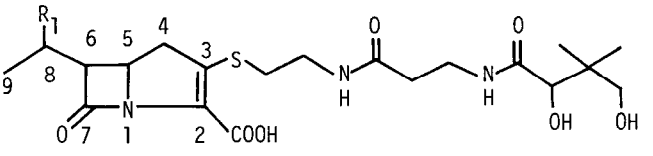
ABSTRACT The structures of carbapenems OA-6129A, B_1, B_2 and C were determined by spectroscopy and chemical transformation

In the course of our extensive work on carbapenems, new compounds OA-6129A(1), B_1(2), B_2(3) and C(4)[1] have been isolated as sodium salts from *Streptmyces sp.* OA-6129 These compounds showed potent antimicrobial activity against Gram-positive and -negative bacteria[2] and inhibitory activity to β-lactamases In this communication, we report the structure determination of the antibiotics by means of spectroscopic analysis, chemical degradation and direct derivation from carbapenem PS-5

The u v and i r spectra of the four compounds strongly suggested that they had a carbapenem skeleton, a u v absorption maximum at 300 nm and i r absorption bands at 1750-1760(β-lactam carbonyl), 1660(amide carbonyl) and 1600 cm^{-1}(carboxylate anion) were very characteristic of the carbapenem structure[3,4] The presence of a pantothenyl group in these

Fig 1



		R_1	C-5	C-6	C-8	Pantoyl group
OA-6129A	(1)	H	*R*	*R*	—	*R*
OA-6129B_1	(2)	OH	*R*	*R*	*	*R*
OA-6129B_2	(3)	OH	*R*	*S*	*S*	*R*
OA-6129C	(4)	OSO_3H	*R*	*R*	*	*R*

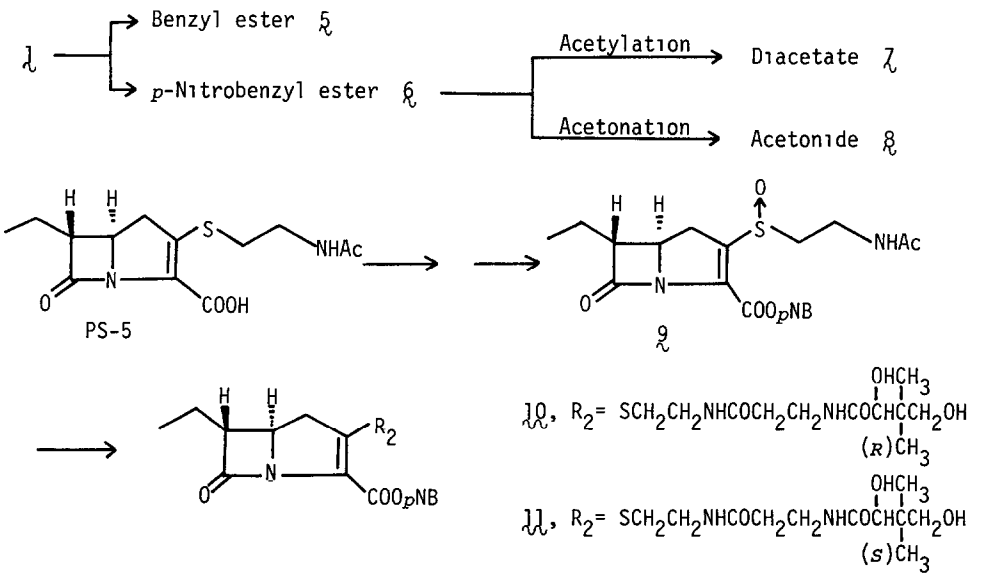
* not determined

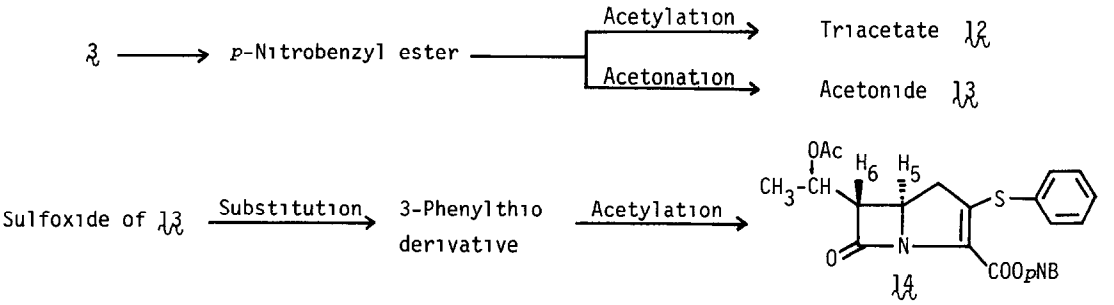
compounds was apparent from their n m r spectra[5] (two singlets attributed to geminal dimethyls at δ 0 86-0 92 and a triplet assigned to α-methylene attached to carbonyl at δ 2 45-2 48) and the formation of the corresponding acetonides and acetates These findings clearly indicated that the four antibiotics had closely related structures including a carbapenem skeleton and a pantothenyl group

The molecular formula of 1 $C_{20}H_{31}N_3O_7S$ was deduced from the FD-MS spectrum [m/z 677 (M+1)] of the diacetate 7 Similarly to both cases of PS-5[3] and N-acetylthienamycin,[4] the characteristic u v absorption maximum of the sodium salt of 1 at 300 nm shifted to 318-319 nm by esterification,[6,7] indicating that 1 had the same chromophore as PS-5 Acid-catalyzed hydrolysis of 1 (6N-HCl, 115°C) gave β-alanine The mass spectrum of benzyl ester 5[7] revealed a strong peak at m/z 259 which showed the presence of cysteamine at C-3,[8] as found in PS-5 benzyl ester[3] Pantoate, β-alanine and cysteamine, taken together, clearly supported the presence of a pantetheinyl group at C-3[8] in 1. The side chain at C-6 was concluded to be ethyl by comparing the n m r signals of 1 and PS-5 The *trans* configuration of the protons at C-5 and C-6 was confirmed by analysis of the lanthanide-induced n m r spectrum of the acetonide 8, as the proton at C-5 appeared as a doublet of triplets ($J_{5,6}$=3 0, $J_{4,5}$= 9 0 Hz) These data allowed us to conclude that 1 was a derivative of PS-5 which had pantetheine at C-3[8] instead of N-acetylcysteamine

The structure of 1 was established by direct conversion of PS-5 to 1 according to a method reported previously[9] S-Oxide of *p*-nitrobenzyl PS-5 9 that was prepared from PS-5 by esterification and peracid oxidation was treated with (*R*)- and (*S*)-pantetheines to give 10 and 11, respectively. Product 10 was identical with 6[6] in every respect[10] Consequently, the structure of 1 was unambiguously determined to be (5*R*,6*R*)-6-ethyl-7-oxo-3-[(*R*)-pantetheinyl]-1-azabicyclo[3 2 0]hept-2-ene-2-carboxylic acid

The molecular formula of 3 $C_{20}H_{31}N_3O_8S$ was also deduced from the FD-MS [m/z 735(M+1)] of the triacetate 12 In the n m r spectrum[5] of 3 1-hydroxyethyl instead of C-6 ethyl in





1 was observed, while the remaining signals resembled very closely those of 1 The *trans* relationship between H-5 and H-6 was obvious from a coupling constant of $J_{5,6}$=3 0 Hz in the n m r spectrum of the acetate 14[11] which was derived from the acetonide 13 by thiophenol replacement[9] *via* the S-oxide The absolute configuration at C-8 bearing a hydroxy group was determined to be *S* by the Mosher's n m r -configuration correlation method[12] using the (*R*)-(+)- and (*S*)-(-)-MTPA esters[4,13] of 13 Thus, the structure of 3 was (5,6-*trans*)-6-[(*S*)-1-hydroxyethyl]-7-oxo-3-pantetheinyl-1-azabicyclo[3 2 0]hept-2-ene-2-carboxylic acid

Compound 2, a minor component, was almost identical with 3 in n m r[5], except for the signal of H-6 at δ 3 60 As the observed chemical shift and coupling constants were in good accordance with those reported for epithienamycin A,[14] 2 was concluded to be an epimer of 3 at C-6

Elemental analysis gave a molecular formula of $C_{20}H_{31}N_3O_{11}S_2$ for 4[15] The presence of a hydroxysulfonyloxy group in 4 was indicated by an i r absorption band at 1220-1250 cm^{-1} and relatively high mobility on high voltage paper electrophoresis[16] Comparison of the n m r spectra[5] of 2 and 4 clearly showed that 4 was a sulfate of 2, as both the methyl doublet and the H-8 multiplet shifted down-field in the spectrum of 4 The *cis* configuration between H-5 and H-6 was apparent from a coupling constant of $J_{5,6}$=5 5 Hz Consequently, 4 was decided to be (5,6-*cis*)-6-(1-hydroxysulfonyloxyethyl)-7-oxo-3-pantetheinyl-1-azabicyclo[3 2 0]hept-2-ene-2-carboxylic acid

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REFERENCES AND NOTES

1 M Okabe, S Azuma, I Kojima, R Okamoto, K Kouno, Y Fukagawa & T Ishikura, submitted for publication in J Antibiotics

2 M Sakamoto, I Kojima, M Okabe, Y Fukagawa & T Ishikura, submitted for publication in J Antibiotics

3 K Yamamoto, T Yoshioka, Y Kato, N Shibamoto, K Okamura, Y Shimauchi & T Ishikura, J Antibiotics 33, 796(1980)

4 G Albers-Schonberg, B H Arison, O.D Hensens, J Hirshfield, K Hoogsteen, E A Kaczka, R E Rhodes, J S Kahan, F M Kahan, R W Ratcliffe, E Walton, L J Ruswinkle, R B Morin & B G Christensen, J.Amer Chem Soc 100, 6491(1978)

5 NMR(D_2O,DSS) δ ppm 1, 0 89(3H,s), 0 92(3H,s), 1 00(3H,t,*J*=7 5Hz), 1 60-2 00(2H,m), 2 48 (2H,t,*J*=6 5Hz), 2 80-3 65(11H,m), 3 95(2H,m). 2, 0 86(3H,s), 0 89(3H,s), 1 33(3H,d, *J*=6 0Hz), 2 47(2H,t,*J*=6 5Hz), 2 75-3 55(10H,m), 3 60(1H,dd,*J*=5 0&10 0Hz), 3 93(1H,s),

3 95-4 40(2H,m) 3, 0 87(3H,s), 0 92(3H,s), 1 28(3H,d,J=7 0Hz), 2 45(2H,t,J=6 5Hz), 2 75-3 60(11H,m), 3 94(1H,s), 3 95-4 35(2H,m) 4, 0 86(3H,s), 0 89(3H,s), 1 49(3H,d, J=6.5Hz), 2 47(2H,t,J=7.0Hz), 2 70-3 60(10H,m), 3 83(1H,dd,J=5 5&9 5Hz), 3 94(1H,s), 4 10-4 43(1H,m), 4 78(1H,dq,J=9 5&6 5Hz)

6 6, UV λmax nm(ε) 319(8400), 270(10500), IR νmax cm^{-1} 1770(β-lactam), 1700(ester), 1665 (amide), NMR(CD_2Cl_2,TMS) δ ppm 0.87(3H,s), 0.95(3H,s), 1 04(3H,t,J=7 5Hz), 1 50-2 20 (3H,m), 2 40(2H,t,J=6 5Hz), 2 80-3 70(12H,m), 3 94(2H,m), 4 17(1H,br), 5 19(1H,d, J=14 0Hz), 5 45(1H,d,J=14 0Hz), 6 74(1H,br), 7 63(2H,d,J=9 0Hz), 8 18(2H,d,J=9 0Hz)

7 5, UV λmax nm(ε) 318(7400), MS(m/z) 259[]$^+$, 242

8 In this paper we employed the numbering system as shown in Fig 1

9 K Yamamoto, T Yoshioka, Y Kato, K Isshiki, M Nishino, F Nakamura, Y Shimauchi & T Ishikura, Tetrahedron Lett 23, 897(1982)

10. $[\alpha]_D^{24}$ (c 1 0,CH_2Cl_2), 6 +52 4°, 10 +51 7°, 11 +30 3°

11 14, NMR($CDCl_3$,TMS) δ ppm 1 36(3H,d,J=6 5Hz), 2 03(3H,s), 2 66(2H,d,J=9 0Hz), 3 28 (1H,dd,J=3 0&4 5Hz), 3 92(1H,dt,J=3 0&9 0Hz), 4 95-5 40(2H,m), 5 50(1H,d,J=14 0Hz), 7 20-7 57(5H,m), 7 60(2H,d,J=9 0Hz), 8 15(2H,d,J=9 0Hz)

12 J A.Dale & H S Mosher, J Amer Chem Soc 95, 512(1973)

13 NMR($CDCl_3$,TMS) δ ppm, (R)-(+)-MTPA ester 0 94(3H,s), 1 00(3H s), 1 39(3H,s), 1 43(3H,s), 1 52(3H,d,J=7 5Hz,H-9), 2 43(2H,t,J=6 5Hz), 2 80-3 75(14H,m), 3 96(1H,m), 4 03(1H,s), 5 22(1H,d,J=14 0Hz), 5 40(1H,m), 5 41(1H,d,J=14 0Hz), 6 48(1H,br), 6 87(1H,br), 7 25-7 64(7H,m), 8 12(2H,d,J=8 0Hz) (S)-(-)-MTPA ester 0 93(3H,s), 1 00(3H,s), 1 39(3H,s), 1 42(3H,s), 1 42(3H,d,J=7 5Hz,H-9), 2 43(2H,t,J=6 5Hz), 2 68-3 85(14H,m), 4 03(1H,s), 4 05(1H,m), 5 22(1H,d,J=14 0Hz), 5 43(1H,d,J=14 0Hz), 5 45(1H,m), 6 47(1H,br), 6 90 (1H,br), 7 22-7 70(7H,m), 8 12(2H,d,J=8 0Hz)

14 P J Cassidy, G Albers-Schonberg, R T Goegelman, T Miller, B Arison, E O Stapley & J Birnbaum, J Antibiotics 34, 637(1981)

15 Elemental analysis ($C_{20}H_{29}N_3O_{11}S_2Na_2$ H_2O), Found C 39 18%, H 4 91%, N 6 86%, S 10 08% Calcd C 39 02%, H 5 08%, N 6 83%, S 10 42%

16 Rm PS-5, 1 69 for 4, 0 67 for 1, 2 & 3 at pH 8 4

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