Communication to the editor

A NEW ANTIBIOTIC, KINAMYCIN[1]

Sir:

An orange crystalline antibiotic, kinamycin was isolated from the culture broth of *Streptomyces murayamaensis* sp. nov. HATA *et* OHTANI.

This strain resembles *Nocardia* in several morphological properties. It produces no aerial mycelium on various agar media. Colonies do not penetrate into the media, and can easily be taken off with a platinum loop. The filamentous, vegetative mycelium divided into a rod form after 2 or 3 days culture. Moreover on the infrared absorption spectrum the mycelium had no absorption at 2930, 1700~1650, and 1460 cm^{-1} as is characteristic of *Nocardia*[2].

For antibiotic production, the medium used was as follows: glucose 2.0 %, soy bean meal 2.0 % and sodium chloride 0.3 %. The pH of medium was adjusted to 8.0 with sodium hydroxide before sterilization. Antimicrobial activity attained a maximum after cultivation for 40 hours in a jar fermenter.

The antibiotics in the culture filtrate was extracted with benzene. After removal of the solvent, residual extract was chromatographed over silicic acid treated with 2 % sulfuric acid, and developed with a mixed solvent of benzene-acetone (20 : 0~7). The active eluate was combined and concentrated to dryness under reduced pressure. On the thin-layer chromatography using Kieselgel G and a mixed solvent of chloroform-ethyl acetate (3 : 2), the residual crude material showed four active components, named kinamycin A (Rf : 0.89), B (0.82), C (0.47), and D (0.39). The crude kinamycin complex was submitted to silicic acid column chromatography using a mixed solvent of chloroform-ethyl acetate (5 : 0~1). The components A, C, and D were crystallized from ethyl acetate and B from chloroform.

Physical and chemical properties of four components are shown in Table 1. The IR (1650~1600 cm^{-1}) (Fig. 1) and UV spectra (240~450 mμ) indicated that each component

Table 1. Physicochemical properties of kinamycins A, B, C, and D

		A	B	C	D
m. p. (°C)		139~142 (dec.)	190~193 (dec.)	150~153 (dec.)	170~175 (dec.)
$[\alpha]_D^{25}$ (c 1, $CHCl_3$)		−60	−48	−24	−37
Elementary analysis found	C	52.52 %	60.58 %	58.40 %	56.78 %
	H	4.39	3.91	4.55	3.90
	N	4.10	4.84	4.92	6.55
	O	38.89	30.67	32.13	32.77
Molecular weight	Mass M^+ (M/e)	436	—	496	—
	RAST method	445	401	494	458
Molecular formular		$C_{19}H_{20}N_2O_{10}$	$C_{20}H_{14}N_2O_7$	$C_{24}H_{20}N_2O_{10}$	$C_{21}H_{18}N_2O_8$
UV (MeOH) mμ ($E^{1\%}_{1cm}$)		248 (660) 279 (305) 396 (sh) (178) 443 (180)	248 (760) 277 (sh) (400) 300 (sh) (230) 400 (225)	246 (528) 275 (sh) (290) 370 (144) 388 (168) 448 (176)	257 (576) 278 (sh) (308) 410 (184) 464 (184)
IR (KBr) cm^{-1}		3400 1370 2155 1250 1736 1225 1615 1155 1450 1075	3360 1449 2155 1372 1738 1230 1616 1153 1481 1076	3510 1625 2900 1486 2155 1460 1745 1370 1663 1225 1155 1075	3600 1477 2900 1455 2155 1370 1733 1225 1640 1155 1075

Table 2. Antimicrobial spectra of kinamycins A, B, C, and D

Test organism	MIC (mcg/ml)				
	A	B	C	D	
Bacillus subtilis PCI-219	0.024	0.012	0.19	0.012	*
Bacillus anthrasis	0.19	0.012	0.19	0.024	*
Staphylococcus aureus FDA 209P	0.78	0.012	0.78	0.024	*
Staphylococcus aureus SM-(R)	1.56	0.006	0.39	0.024	*
Staphylococcus albus	0.024	0.012	0.39	0.024	*
Streptococcus hemolyticus (Cook)	100	12.5	12.5	50	**
Streptococcus hemolyticus (NY-5)	1.56	3.15	12.5	12.5	**
Streptococcus hemolyticus (S-8)	6.25	6.25	12.5	12.5	**
Mycobacterium 607	25	6.25	6.25	6.25	*
Escherichia coli NIHJ	100	3.12	100	12.5	*
Vibrio comma	100	0.19	25	12.5	*
Vibrio comma Inaba 904	50	0.09			*
Klebsiella pneumoniae	>100	12.5	100	25	*
Pseudomonas aeruginosa P-1	>100	>100	>100	>100	*
Pseudomonas aeruginosa P-2	>100	>100	>100	>100	*
Salmonella typhosa 901 W	>100	6.25	>100	12.5	*
Salmonella paratyphi A	>100	>100	>100	25	*
Salmonella enteritidis gartnerii	>100	>100	>100	100	*
Shigella dysenteriae	>100	25	>100	25	*
Proteus vulgaris OX-19	>100	12.5	>100	6.25	*
Neisseria gonorrhoeae	50	12.5	>100	50	**
Saccharomyces sake	>100	>100	>100	>100	***
Candida albicans	>100	>100	>100	>100	***
Aspergillus niger	>100	>100	>100	>100	***
Penicillium chrysogenum Q-176	50	>100	>100	12.5	***

Inoculum size: One loopful of test organism
Medium: * Nutrient agar ** Blood agar *** Potato agar

has a quinone group[3]. Each component was soluble in methanol, acetone, chloroform, and ethyl acetate slightly soluble in ethyl ether and benzene, and insoluble in water and *n*-hexane, and gave a positive color reaction with ferric chloride, LIEBERMANN, rhodunin and ammonia, and 1-phenyl-3-methyl pyrazol-5-one[4], and negative reactions with TOLLENS, FEHLING, ninhydrin, MILLON, nitroprusside, and 2-aminothiophenyl reagents.

As shown in Table 2, kinamycins A, B, C, and D are mainly active against Gram-positive bacteria, and less active against Gram-negative bacteria. The acute toxicity (LD_{50}) of four components in mice is about 30~40 mg/kg each, intravenously.

Several quinone antibiotics, such as frenolicin[5], aquayamycin[6], tetrangomycin[7], julimycin[8] and daunomycin[9] were reported. The kinamycins have an infrared absorption maximum at 2155 cm^{-1}, assignable to a nitrile or isonitrile group, and can be differentiated from antibiotics lacking this absorption.

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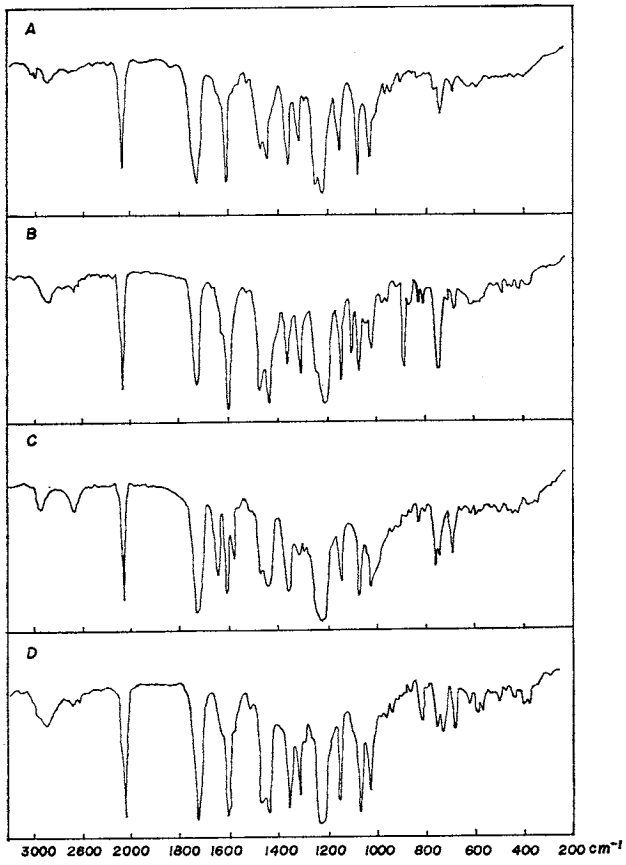
Fig. 1. Infrared spectra of kinamycins A, B, C, and D (KBr)



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