ANTIBIOTICS PRODUCED BY HYDROCARBON-UTILIZING ACTINOMYCETES

Sir:

Recently, hydrocarbons and non-sugar carbohydrates such as alcohols and their esters have been considered as a sole carbon source for the growth of microorganisms, and actually been utilized in some fermentation industries. In the field of antibiotic fermentation, pyocyanine[1], 1-phenazine carboxylic acid[2,3], pyoluteorin, its relative compounds[4], fluopsins[5,6] and *p*-nitrophenylserinol derivatives[7] were isolated from the cultured broth of *n*-paraffin-utilizing bacteria. However, there have not been any reports of the production of any antibiotics by hydrocarbon-utilizing actinomycetes.

Therefore, we have made attempts to search hydrocarbon-utilizing actinomycetes, and obtained some strains belonging to *Streptomyces* and capable of utilizing methyl acetate, *n*-paraffins and so on. Of these strains, No. 81 and No. 351 were selected in the present investigation concerning the production of antibiotics.

The organisms were cultivated in a basal medium composed of 0.5 % $(NH_4)_2SO_4$, 0.2 % K_2HPO_4, 0.1 % KH_2PO_4, 0.05 % $MgSO_4 \cdot 7H_2O$ and 6 % (v/v) vitamin solution*, pH 7.0. Three percent NP-200**, light gas oil***, methyl acetate, methanol or sodium acetate was added as the sole carbon source to the basal medium. Cultivation was carried out on a reciprocal shaker at 12°C and at 28°C.

Strain No. 81, which resembles *Streptomyces globisporus*, is a facultative psychrophile isolated from a soil sample collected in Kyoto. This strain was found to produce a new peptide antibiotic, cryomycin[8,9], at low temperature, and a water-soluble basic antibiotic, tentatively named M-81[10], at moderate temperature, from hydrocarbons, esters and so on (Table 1). The maximum production of cryomycin was achieved after 10 days of cultivation at 12°C, and that of M-81 after 5~6 days at 28°C.

Strain No. 351, which resembles *Streptomyces phaeochromogenus*, is a psychrophile isolated from a soil sample collected in Hiroshima. This strain produced certain water-insoluble antibiotics from methyl acetate, NP-200 and light gas oil at 28°C (Table 1). Preliminary investigations revealed that the antibiotics were quite similar to nonactin and its related compounds. The maximum production was achieved after about 5 days at 28°C.

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Table 1. Antibiotic production of *Streptomyces* sp. No. 81 and No. 351 from various carbon sources

Carbon source	*St.* sp. No. 81		*St.* sp. No. 351
	Cryomycin (12°C, 10 days)	M-81 (28°C, 5 days)	(28°C, 5 days)
NP-200	20 mcg/ml	150 units/ml*	trace
Light gas oil	20	300	<100 mcg/ml
Methyl acetate	70	500	<100
Sodium acetate	0	100	
Methanol	35	200	
Control (no carbon source)	0	0	0

* One unit was designated as the minimal inhibitory concentration against *Serratia polymuthicum* IFO 3055 at pH 7.0.

* Vitamin solution: Thiamine-HCl 2 mg, riboflavin 2 mg, nicotinic acid 4 mg, biotin 16 mcg, Ca-pantothenate 2 mg, *p*-aminobenzoic acid 0.4 mg, folic acid 0.04 mg and pyridoxol 62.5 mcg in a total volume of 250 ml.

** NP-200: *n*-paraffin (C_{14} 68.1 %, C_{15} 29.9 %)

*** Light gas oil: *n*-paraffins 40.1 % and non *n*-paraffins 58.8 %.

NP-200 and light gas oil were supplied as gifts from Mr. K. Minami, Maruzen Petroleum Industry Co., Ltd.

References

1) Lee, E.G.H. & C. C. Walden : Biosynthesis of pyocyanine by a paraffin hydrocarbon-oxydizing strain of *Pseudomonas aeruginosa*. Appl. Microbiol. 17 : 520～523, 1969
2) Higashihara, T. & A. Sato : Formation of 1-phenazine carboxylic acid from hydrocarbons by microorganisms. J. Ferment. Technol. 48 : 73～78, 1970
3) Ogata, K.; K. Minami & Y. Tani : Formation of 1-phenazine carboxylic acid and oxychlororaphine from hydrocarbons by a microorganism. J. Ferment. Technol. 49 : 925～934, 1971 (in Japanese)
4) Omori, T. & K. Yamada : Formation of antibacterial substances from hydrocarbons by microorganisms. p. 11, Abstracts of papers, The Annual Meeting of Agricultural Chemical Society of Japan, Sendai, 1972
5) Itoh, S.; K. Inuzuka & T. Suzuki : New antibiotics produced by bacteria grown on *n*-paraffin (Mixtures of C_{12}, C_{13} and C_{14} fractions). J. Antibiotics 23 : 542～545, 1970
6) Shirahata, K.; T. Deguchi, T. Hayashi, I. Matsubara & T. Suzuki : The structures of fluopsins C and F. J. Antibiotics 23 : 546～550, 1970
7) Suzuki, T.; K. Shirahata, H. Honda, T. Hayashi & R. Katsumata : Production of *p*-nitrophenylserinol derivatives by *n*-paraffin-growing bacterium. p. 200, Abstracts of papers, The IV th International Fermentation Symposium, Kyoto, 1972
8) Ogata, K.; N. Yoshida, M. Ohsugi & Y. Tani : Studies on antibiotics produced by psychrophilic microorganisms. I. Production of antibiotics by a psychrophile, *Streptomyces* sp. No. 81. Agr. Biol. Chem. 35 : 79～85, 1971
9) Yoshida, N.; Y. Hachiya, Y. Tani & K. Ogata : Studies on antibiotics produced by psychrophiles. II. Cryomycin, a new antibiotic. p. 119, Abstracts of papers, The Annual Meeting of Agricultural Chemical Society of Japan, Tokyo, 1971, J. Antibiotics (in preparation)
10) Yoshida, N.; S. Hayashi, Y. Tani & K. Ogata : Studies on antibiotics produced by a psychrophile, *Streptomyces* sp. No. 81, at moderate temperature. p. 15, Abstracts of papers, The Annual Meeting of Agricultural Chemical Society of Japan, Sendai, 1972, J. Antibiotics (in preparation)