SHORT COMMUNICATIONS

*Glumamycin, A New Peptide-type Antibiotic**

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A Streptomyces strain isolated in March, 1954 from a sample collected in Osaka Prefecture resembled the Zaomycin producing strain, S. zaomyceticus[1], in morphological and cultural characteristics and the utilization of carbon source, and therefore it was named S. zaomyceticus No. 7548 tentatively. The strain, when cultivated on a medium containing 3.0% of starch, 1.0% of polypepton, 0.2% of potassium phosphate 0.3% of soybean oil, and 2.0% of rice bran for 96 hr., produced an antibiotic, but the potency lowered abruptly thereafter.

The antibiotic inhibited the growth of Gram-positive bacteria such as Micrococcus aureus 209p and B. subtilis and was especially active against the bacteria resistant to known antibiotics. Although this antibiotic was similar to Amphomycin[2,3], Zaomycin[1] and Crystallomycin[4,5], it differed from them clearly. The authors, having found it to be a new antibiotic, named it Glumamycin and investigated its constituents.

The antibiotic was isolated pure from the broth by Craig's counter current distribution method 300 plates; chloroform- methanol-0.02 N hydrochloric acid (2:2:1), and it gave only one spot in paper chromatography and paper ionophoresis at various pH's. M. p. 230°C (decomp.). Found: C, 51.71; H, 7.20; N, 12.99%. From its dissociation constant, the molecular weight was estimated to be about 1800. The antibiotic had its isoelectric point at pH 3.4, and though it showed the typical infrared-spectrum characteristic of peptides, it exhibited no peculiar absorption in the ultraviolet-region.

The antibiotic was not hydrolized by any hydrolyrase such as pepsin, trypsin and carboxypeptidase, but it was completely hydrolized by heating with 6 N hydrochloric acid at 110°C for 20 hr. When the hydrolysate was subjected to two dimensional paper chromatography *n*-butanol-acetic acid-water (4:1:5) or 80% phenol containing 0.5 N ammonia, six amino acids were detected, four of which were aspartic acid, glycine, valine and proline and the other were unknown. Of the two unknown amino acids, one was a basic amino acid differing from lysine, histidine and arginine on paper chromatogram, and the other was a neutral amino acid which was characterized by a violet blue color with ninhydrin and a blue color with isatin. The six amino acids were separated from each other in crystalline form when chromatographed first on ion-exchange resin (IR-4B, Dowex 50×8) and then on cellulose powder.

After recrystallization from 75% methanol, the unknown basic amino acid showed m. p. 202°C (decomp.), $[\alpha]_D^{22}=+16°$ (c, 1 : H_2O), Pk <2, 6.6, 9.6, and gave the values of 1.0 mol. of C–CH_3 (Kuhn-Roth) and 1.8 mol. of amino nitrogen (Van Slyke), and it was different from authentic α,γ-diaminobutyric acid on paper chromatogram. From these facts it was presumed to be α,β-diaminobutyric acid. On the other hand, the unknown neutral amino acid, when recrystallized from methanol·ether, melted at 240°C (decomp.), showed $[\alpha]_D^{21}=+10°$ (c, 1 : H_2O) and accorded with $C_6H_{11}O_2N$·HCl, but no amino nitrogen (Van Slyke) and C–CH_3 (Kuhn-Roth) were detected. This compound was in complete agreement with synthetic DL-pipecolic acid in paper chromatography with various solvent systems, and the optical rotations of its hydrochloride and tartarate were $[\alpha]_D^{25}=+10°$ (c, 1 : H_2O) and $[\alpha]_D^{25}=+20°$ (c, 1 : H_2O), respectively. As the sign of optical activity of the neutral amino acid was opposite to that of the natural pipecolic acid it belongs to the D-series; the aspartic acid, valine, and proline derived from the antibiotic belong to the L-series.

The hydrolysate mentioned before contained about 10% of a substance soluble in ether, which was found to ba a monobasic unsaturated fatty acid, b. p. 139°C (1 mmHg), $[\alpha]_D^{21}=+4°$

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1) Y. Hinuma, *J. Antibiotics*, ser. A 7, 134 (1954).
2) B. Heinemann, M. A. Kaplan, et al., *Antib. & Chemoth.*, 3, 1239 (1953).
3) G. Giolitti, G. Corti et al., *Giorn. Microbiol.*, 3, 70 (1957).
4) G. F. Game et al., *Antibiotiki*, 2, (6) 14 (1957).
5) N. N. Lomakina and M. G. Branzhnikova, *Biokhimiya* 24, 425 (1959).

(c, 1 : C_2H_5OH), $C_{13}H_{24}O_2$. Oxidation of the substance with ozone afforded a compound $C_9H_{18}O_2$, b. p. 154°C (25 mmHg), $[\alpha]_D^{21} = +12°$ (c. 1 : C_2H_5OH), and from this and other results, a 4-isotridecenoic acid structure, $C_8H_{17}CH=CHC_2H_4$–COOH, was assigned to the unsaturated fatty acid.

Thus Glumamycin was assumed to be composed of 4-isotridecenoic acid, L-aspartic acid, glycine, L-valine, L-proline, D-pipecolic acid, and α, β-diaminobutyric acid combined through peptide linkages. Full details of this work will be reported shortly.

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