

KF77-AG6, AN ANTIPAIN-RELATED METABOLITE

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Recently we reported¹⁾ the isolation and structural elucidation of a SAKAGUCHI-positive metabolite of *Streptomyces*, found in the course of chemical screening of microbial metabolites. The compound proved to be identical to antipain²⁾ discovered as a protease inhibitor. In the present paper, we wish to report the isolation of another SAKAGUCHI-positive metabolite, KF77-AG6, which corresponds to the ureylene group-containing part of antipain, *i.e.* [(S)-1-carboxy-2-phenylethyl]carbamoyl-L-arginine³⁾. KF77-AG6 is produced by a strain of *Streptomyces filipinensis*³⁾ (the Laboratory No. KF77-AG6), from which antipain was not produced.

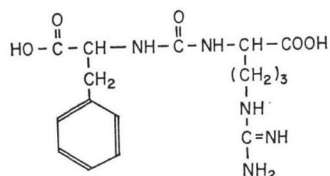
A part of the biological effect of KF77-AG6 (described as AN) has already been reported³⁾.

Experimental

Fermentation was carried out in a medium (pH 7.0) consisting of glycerol 2.5 %, yeast extract 1.0 %, peptone 0.5 %, CaCO₃ 0.3 %, NaCl 0.2 %, MgSO₄·7H₂O 0.05 % and K₂HPO₄ 0.05 %, at 27°C for 5 days. The broth (pH 8.4) was filtered and the filtrate (4.5 liters) was extracted twice with 1-butanol (2 l×2). The extract was evaporated to give a solid (8.2 g),

* Taxonomy by YOSHIRO OKAMI of the Institute of Microbial Chemistry.

which was chromatographed on a column (4.8×14 cm) of alumina (Woelm acid form, 220 g), and was successively eluted with methanol (500 ml), water (800 ml) and 1 N aqueous ammonia. The ammoniacal fraction (210~300 ml) positive for diacetyl reagent⁴⁾ was evaporated and the resulting solid (2.80 g) was chromatographed on a column (3.7×55 cm) of cellulose ("Avicel", 200 g) with 1-butanol - ethanol - water (3 : 1 : 2). The diacetyl-positive fraction (615~930 ml) was evaporated to give a solid (1.0 g), which was further purified by chromatography on Amberlite CG-50 (H-form 50 ml; 1.7×21 cm) with water, and on Dowex 1×2 (Cl from 20 ml; 1.2×16 cm) with 0.1 N hydrochloric acid. KF77-AG6 was obtained as a monohydrochloride (160 mg): a hygroscopic solid, mp 125~141°C (dec.) (from water-acetone); [α]_D²⁰+5° (c 1.0, water); pK_a 3.2, 4.1, >11; positive for SAKAGUCHI, diacetyl, RYDON-SMITH and WOOD reagents, but negative for ninhydrin.



KF 77-AG6

References

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