
Notes

A NEW ENTEROMYCIN GROUP
ANTIBIOTIC, YN-0165J-A
PRODUCED BY
STREPTOMYCES SP.

HARUMITSU IMAI, KEN-ICHI SUZUKI,
SHUICHI TAKAMURA, SHIGENOBU KADOTA
and MASARU IWANAMI

Bioscience Research Laboratories,
Central Research Laboratories,
Yamanouchi Pharmaceutical Co., Ltd.,
1-1-8, Azusawa, Itabashi-ku,
Tokyo, Japan

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In the course of our screening for new antibiotics, a *Streptomyces* strain YN-0165J isolated from a soil sample collected at Omaezaki in Shizuoka Prefecture, Japan, was found to produce a new antibiotic.

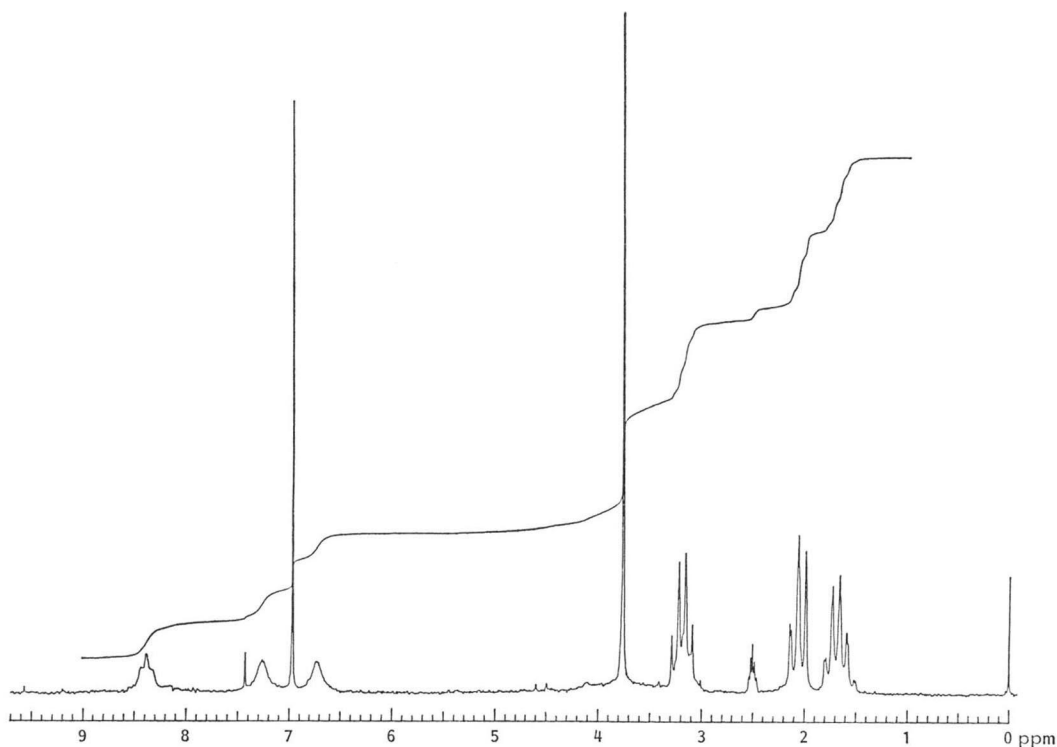
The strain YN-0165J is classified in the genus

Streptomyces on the basis of the following characteristics: Color of mature sporulated aerial mycerium is in the blue-color series; mature spore chains showing predominantly hooks, loops or incomplete spirals is section Retinaculum-Apertum; spore surface is warty; formation of melanoid pigment is negative. Analysis of whole cell hydrolysate of the strain YN-0165J showed that it contained LL-diaminopimelic acid and glycine.

The strain was cultured in 500-ml Erlenmeyer flasks containing 60 ml of a medium consisted of potato starch 3.0%, soybean meal 1.5%, yeast extract 0.2%, corn steep liquor 0.5%, $MgSO_4 \cdot 7H_2O$ 0.05%, NaCl 0.3% and $CoCl_2 \cdot 6H_2O$ 0.001%. The medium was adjusted to pH 7.0 before sterilization. The strain was cultured at 27°C for 72 hours on a rotary shaker. The antibiotic activity was monitored by paper disk assay using *Escherichia coli* K-12 as a test organism.

The clarified broth (10 liters) was applied to a Diaion HP-20 resin column. After washing

Fig. 1. 1H NMR spectrum of YN-0165J-A (100 MHz, $DMSO-d_6$).



with water, the antibiotic was eluted with 50% acetone. The active fractions were collected and concentrated to dryness. The solid residue (10.75 g) was dissolved with CHCl_3 - MeOH, 4:1 and filtered. The filtered solution was concentrated to a small volume, and then chromatographed on silica gel (120 g) eluting with CHCl_3 - MeOH, 9:1. The active fractions were collected and concentrated to afford a white powder. The powder was crystallized from ethyl acetate to give white crystals (267 mg).

The physico-chemical properties of YN-0165J-A are as follows: MP 120~121°C (dec); high resolution CI-MS m/z 204.099 (M+H, $\text{C}_7\text{H}_{14}\text{N}_3\text{O}_4$), 129.066 ($\text{C}_5\text{H}_9\text{N}_2\text{O}_2$); color reactions, positive to 0.5% KMnO_4 and ninhydrin, negative to FeCl_3 and Dragendorff; UV $\lambda_{\text{max}}^{\text{MeOH}}$ nm (ϵ) 252 (14,840); IR (KBr) cm^{-1} 3380, 1650, 1590, 1530, 1430, 1240, 1100 and 1000; ^1H NMR (100 MHz, $\text{DMSO}-d_6$) as shown in Fig. 1; ^{13}C NMR (D_2O) δ 181.7, 164.4, 144.3, 58.1, 41.6, 35.3 and 27.5; Anal Calcd for $\text{C}_7\text{H}_{13}\text{N}_3\text{O}_4$: C 41.38, H 6.45, N 20.68, Found: C 41.42, H 6.68, N 20.64.

From the results described above, it is considered that antibiotic YN-0165J-A is classified in enteromycin group antibiotics such as enteromycin¹⁾, enteromycin carboxamide²⁾, thermycetin³⁾, RP-7080⁴⁾, U-15774⁵⁾, U-22956⁵⁾ and 19A⁶⁾. However, the physico-chemical properties of YN-0165J-A are different from those of the above antibiotics in this group. The analytical and spectroscopic data of YN-0165J-A indicated above, suggested that the structure was 4-[2-(*N*-oxidemethoxyimino)acetamide]butylamide, as shown in Fig. 2. Consequently, YN-0165J-A is considered to be a new antibiotic. The antimicrobial activity of YN-0165J-A is shown in Table 1.

Fig. 2. Chemical structure of YN-0165J-A.



Table 1. Antimicrobial spectrum of YN-0165J-A.

Test organism	MIC ($\mu\text{g/ml}$)
<i>Bacillus subtilis</i> ATCC 6633	25
<i>Micrococcus luteus</i> ATCC 9341	25
<i>Staphylococcus aureus</i> Smith	50
<i>Escherichia coli</i> NIHJ	12.5
<i>Morganella morganii</i> IID 602	25
<i>Enterobacter cloacae</i> 963	100
<i>Pseudomonas aeruginosa</i> NCTC 10490	100

The MIC were determined by a serial agar dilution method with Mueller-Hinton medium.

Inoculum size; 10^6 cfu/ml.

Acknowledgments

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