

Tryptophan Moiety of Rufomycin Homologs^{*1,2}

Amino acid sequences of rufomycin homologs have been shown in the previous paper,¹⁾ and also we suggested the presence of a substituent in the tryptophan moiety. While, T. Takita^{2,3)} isolated a tryptophan derivative from the acidic hydrolysate of ilamycin B₁^{*3} and established the structure of it as II, and also indicated the structure of ilamycin B₁.

The present communication deals with the structure of tryptophan derivatives obtained from rufomycin A and rufomycin B₁. Also, the structure of rufomycin B₁ is described.

Degradation of rufomycin B₁ with a saturated barium hydroxide solution at 100° for 96 hr. yielded a new tryptophan derivative (III), UV : $\lambda_{\text{max}}^{80\% \text{ EtOH}}$ 282.5 m μ , IR $\nu_{\text{max}}^{\text{KBr}}$ cm⁻¹ : 920, 995 (-CH=CH₂), NMR (D₂O) 6 protons of *gem*-methyl (8.3 τ singlet). Catalytic hydrogenation of III over platinum gave a dihydro compound (IV), UV : $\lambda_{\text{max}}^{80\% \text{ EtOH}}$ 285 m μ , and the infrared spectrum of IV showed that the end vinyl group of III was saturated in IV. Treatment of IV with acetic acid-hydrochloric acid (1:1) at 100° for 30 min. gave tryptophan (VI) quantitatively. From these results, the structures of III and IV are estimated as 1-(2-methyl-3-buten-2-yl)tryptophan and 1-(1,1-dimethylpropyl)-tryptophan, respectively. On the other hand, a tryptophan derivative was isolated from a hydrolysate of rufomycin B₁ with acetic acid-hydrochloric acid (1:1) at 100° for 3 hr.³⁾ by the chromatography on an Amberlite CG-50 column with 15% acetic acid. The IR spectrum of the derivative was identical with that of II isolated from ilamycin B₁.³⁾ Peaks for II, III, IV and VI which emerged from a 15 cm. column with pH 5.28 buffer by an amino acid autoanalyser (Beckmann) are obtained respectively at positions about 100 ml., 113 ml., 105 ml. and 47 ml. Structural changes between II, III and IV were confirmed by the amino acid analysis. Treatment with acetic acid-hydrochloric acid (1:1) at 100° for 30 min. or 70% formic acid at 100° for 2.5 hr. converted most of III to II. Substance IV yielded quantitatively VI by the same treatment. Moreover II was recovered without any change by a treatment either with a barium hydroxide solution at 100° for 96 hr. or with acetic acid-hydrochloric acid (1:1) at 100° for 18 hr. These results suggest that III is a original component in rufomycin B₁. To confirm this assumption, hydrorufomycin B₁ (V) obtained by a catalytic reduction of rufomycin B₁ over platinum⁴⁾ was submitted to hydrolysis. Degradation of V with a barium hydroxide solution at 100° for 96 hr. gave IV while treatment of V with acetic acid-hydrochloric acid (1:1) at 100° for 4 hr. yielded L-tryptophan. These results are shown in the Chart 1. Consequently, the structure of rufomycin B₁ is estimated as shown in Fig. 1.

Meanwhile, degradation of rufomycin A and its derivatives gave a new tryptophan derivative. Namely, rufomycin A (VII), dihydrorufomycin A¹⁾ (XI) obtained by the reduction

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^{*2} Part of this paper on rufomycin A and report about tryptophan derivatives from ilamycin and ilamycin B₁ were presented at the 139th meeting of Japan Antibiotics Research Association (J. A. R. A.) by M. Fujino, *et al.* and T. Takita, *et al.*, independently.

^{*3} Rufomycin A and rufomycin B₁ are resemble to ilamycin and ilamycin B₁, respectively.

1) J. Ueyanagi, M. Fujino, T. Kamiya, H. Iwasaki, A. Miyake, S. Tatsuoka : Abstracts of the 7th Symposium on the Chemistry of Natural Products, p. 109 (Oct. 1963).

2) T. Takita : The amino acid sequence of ilamycin and ilamycin B; J. Antibiotics (Japan) Ser. A, **16**, 211 (1963) [The 135th meeting of J. A. R. A. July 27 (1963)].

3) T. Takita, H. Naganawa, K. Maeda, H. Umezawa : A new amino acid from ilamycin B₁ and the structure of ilamycin B₁; J. Antibiotics (Japan) Ser. A, **17**, 90 (1964).

4) J. Ueyanagi, H. Iwasaki, M. Fujino, T. Kamiya, A. Miyake, S. Tatsuoka : Abstracts of the 6th Symposium on the Chemistry of Natural Products, p. 53 (1962).

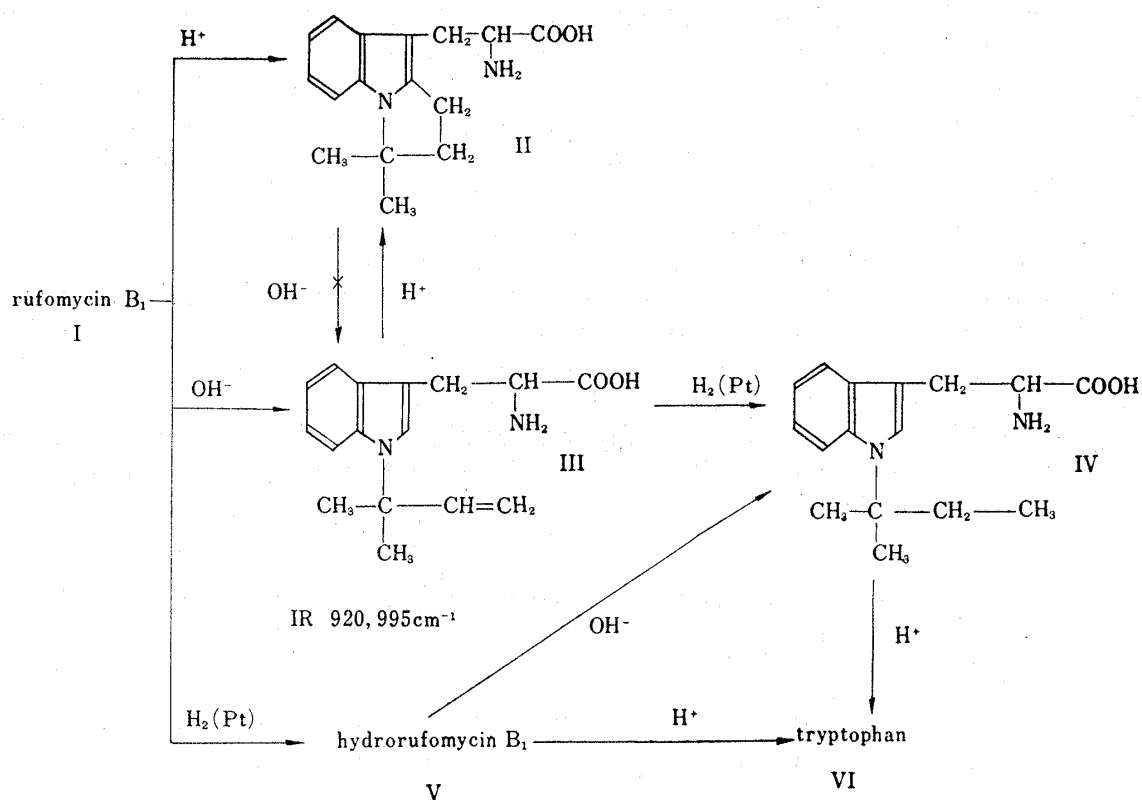
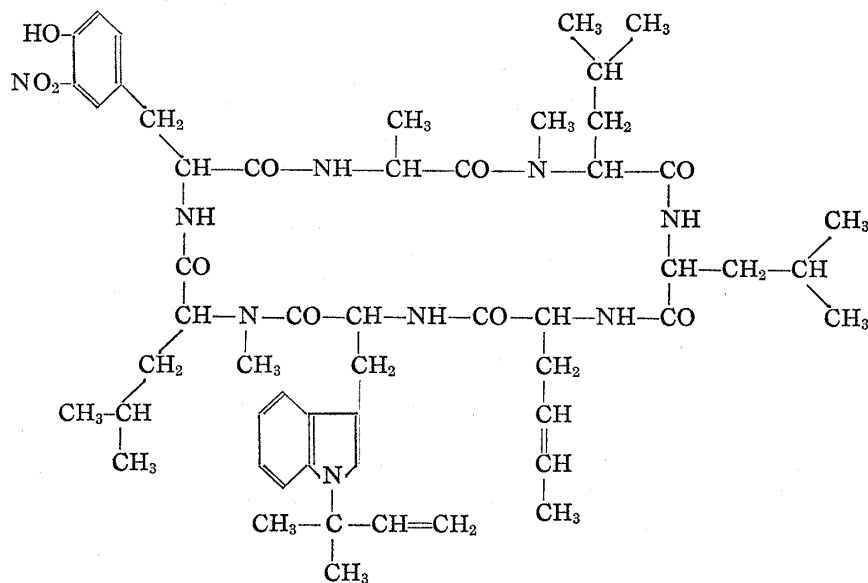


Chart 1.

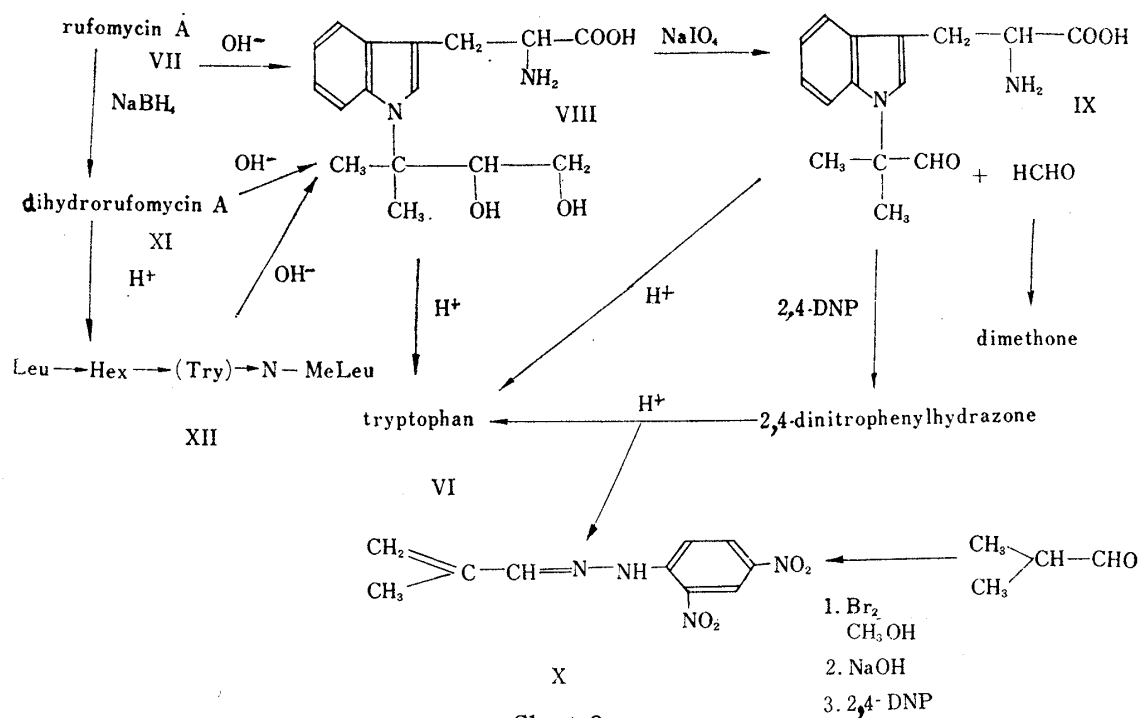
Fig. 1. Structure of Rufomycin B₁

with sodium borohydride⁵⁾ and the partial hydrolysate¹⁾ (XII) were respectively treated with barium hydroxide solutions at 100° for 96 hr. to give a new tryptophan derivative (VIII), *Anal.* Calcd. for $C_{16}H_{22}O_4N_2 \cdot H_2O$ (VIII): C, 59.24; H, 7.46; O, 24.66; N, 8.63. Found: C, 59.51; H, 7.15; O, 24.37; N, 8.40. UV: $\lambda_{\text{max}}^{H_2O}$ 285 m μ , IR: ν_{max}^{KBr} 3300 cm^{-1} (broad: OH), NMR (D_2O) 6 protons of the *gem*-CH₃ (8.31 τ singlet). Treatment of VIII with 2N HCl at 105° for 4 hr. yielded VI, but more drastic conditions with acid converted VIII to a black

5) T. Takita: J. Antibiotics (Japan), Ser. A, 16, 175 (1963).

gummi substance. VIII was also obtained by a hydrolysis of VII with 70% formic acid at 100° for 5~24 hr. To confirm positions of OH-groups, VIII was oxidized with sodium periodate at 5° for 30 min. to formaldehyde (identified as formaldimethone about 40% yield) and IX, UV: $\lambda_{\text{max}}^{\text{H}_2\text{O}}$ 282 m μ ($E_{1\text{cm}}^{1\%}$ 175), IR: $\nu_{\text{max}}^{\text{KBr}}$ 1720 cm⁻¹ (carbonyl), NMR 6 protons of the *gem*-CH₃ (τ 8.29). IX was converted to a 2,4-dinitrophenylhydrazone and treatment of the hydrazone with 2*N* HCl at 100° for 40 min. gave quantitatively VI and a 2,4-dinitrophenylhydrazone of the substituent (X), *Anal.* Calcd. for C₁₀H₁₀O₄N₄: C, 47.61; H, 4.79; N, 22.21. Found: C, 47.80; H, 4.40; N, 21.77. Melting point of X (194° (decomp.)) was not depressed by admixture with the authentic 2,4-dinitrophenylhydrazone of 2-methylacrolein. The UV spectrum of X ($\lambda_{\text{max}}^{\text{CHCl}_3}$ 368 m μ) exhibited characteristics of that of the authentic sample. From these results, the structure of VIII was estimated as 1-(1,1-dimethyl-2,3-dihydroxypropyl)tryptophan.

The reactions described in this communication are shown in the Chart 1 and 2. Further studies on the structure of the original tryptophan moiety of rufomycin A is now under investigations.



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