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IKARUGAMYCIN. I. CHROMOPHORE AND PARTIAL STRUCTURE

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Ikarugamycin, a new antibiotic, was first isolated from the culture broth of Streptomyces phaeochromogenes var. ikaruganensis by K.Jomon and co-workers.[1] In the present paper we wish to report the partial structure I, which contains all the hetero atoms (four oxygen and two nitrogen atoms), for ikarugamycin. This antibiotic has the physicochemical properties as follows.

Ikarugamycin (1): mp 228-229°(dec)[2,3]; $C_{29}H_{38}O_4N_2$; m/e 478 (M^+); $[\alpha]_D$(DMF) +390°; pKa' (67%EtOH) 5.6; $\nu_{max}(CHCl_3)$ 3450, 1700, 1665, 1642, 1580, and 1510 cm^{-1}; λ_{max}(MeOH) 227 and 327 nm (ε 20,700 and 17,300, respectively), λ_{max}(0.1N NaOH-MeOH) 243 and 321 nm (ε 21,400 and 13,300, respectively); δ_{ppm}(pyr-d_5) 0.88(3H, d, J=7Hz), 0.93(3H, t, J=7Hz), 3.6∿4.4(2H, m), 4.1(1H, narrow m), 5.72(1H, br d, J=10Hz), 5.95(1H, br d, J=10Hz), ca. 6.0(1H, m), 6.20(1H, br d, J=12 Hz), 6.94(1H, dd, J=15.6 and 9.5Hz), 7.62(1H, d, J=15.6Hz), 8.7*[4](1H), and 9.6*(1H), δ_{ppm}(DMSO-d_6) 5.6∿6.2(4H, m), 6.64(1H, dd, J=15.0 and 9.5Hz), 6.96(1H, d, J=15.0Hz), 7.79*(1H), and 8.62* (1H). Catalytic hydrogenation of 1 (PtO_2 in EtOH, 1 hr) afforded hexahydroikarugamycin (2); mp 243-244°(dec); $C_{29}H_{44}O_4N_2$; m/e 484 (M^+); pKa'(67%EtOH) 5.1; $\nu_{max}(CHCl_3)$ 3450, 1710, 1662, 1610, and 1520 cm^{-1}; λ_{max}(MeOH) 220 and 280 nm (ε 5,000 and 12,400, respectively), λ_{max}(0.1N NaOH-MeOH) 243 and 279 nm (ε 10,300 and 13,600, respectively); $\delta_{ppm}(CDCl_3)$ 5.86*(1H) and 6.0*(1H).

In the NMR spectrum of 2, the signals corresponding to the olefinic protons in 1 have disappeared, indicating that 1 should have three -CH=CH- groups.† One of them is trans (J=15.6 Hz) and the other two are cis on the basis of their coupling constants (J=12 and 10Hz). In addition, the comparison of UV spectra between 1 (327 nm) and 2 (280 nm) indicates that one double bond must be contained in the main chromophore of 1.

† There is no significant difference between 1 and 2 in the C-Me region, indicating that 1 has no $-CH=CH_2$ group.

The presence of a β-diketone group in 1 and 2 can be explained by their pKa' values and UV spectra coupled with the chemical evidences as follows.

Ferric chloride tests of 1 and 2 were positive (orange-red color). Both 1 and 2 formed yellow-green and blue crystalline copper salts, respectively. Hexahydroikarugamycin (2) reacted with N-methylhydrazine to give a N-methylpyrazole derivative [m/e 494 (M^+)].

On prolonged catalytic hydrogenation of 2 (PtO_2 in EtOH, 24 hr), one carbonyl group of the β-diketone was converted into a methylene group to give deoxyoctahydroikarugamycin (3) [mp 155-157.5°; $C_{29}H_{46}O_3N_2$; m/e 470 (M^+); δ_{ppm}(DMSO-d_6) 7.09*(1H), 7.85*(1H), and 10.32*(1H)], which was further reduced with $LiBH_4$ (in DME, room temp., 24 hr) affording deoxydecahydroikarugamycin (4) [mp 220-222°; $C_{29}H_{48}O_3N_2 \cdot H_2O$; m/e 472 ($M^+$); δ_{ppm}(DMSO-d_6) 5.01*(1H, d, J=5.2Hz), 7.65*(1H), and 7.88*(1H)], This derivative (4) was also obtained directly from 2 according to the similar procedure ($LiBH_4$,in DME, reflux temp., 6 hr).

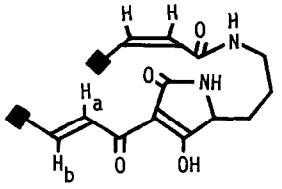
All the NMR spectra of 1~4 show two broad signals due to each one proton, which disappear on addition of D_2O, and their IR spectra have absorption bands near 3450, 1700, 1660, and 1515 cm^{-1}. These findings indicate that ikarugamycin (1) and its derivatives (2~4) must contain two -CO-NH- groups, one of which is an amide or a large-membered ring lactam[5] [ν_{max}($CHCl_3$) 1665 and 1520 cm^{-1} in 4], and the other seems to be a five-membered ring lactam (1695 cm^{-1} in 4).

The presence of a β-keto amide system in 3 was suggested by its spectroscopic properties [pKa'(67%EtOH) 7.8; λ_{max}(MeOH) 220 and sh. 240 nm (ε 6,100 and 4,600, respectively), λ_{max}(0.1N NaOH-MeOH) 222 and 273 nm (ε 3,200 and 9,300, respectively)] and finally confirmed by the formations of an enol acetate [m/e 512 (M^+)] and an enol ether [m/e 484 (M^+)].

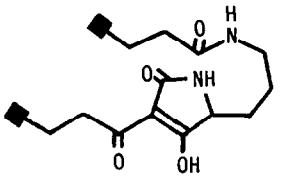
Furthermore, ozonolysis of ikarugamycin (1) in MeOH at -70°, followed by performic acid oxidation, afforded a bicyclic tetracarboxylic acid which was isolated as the tetramethyl ester 5 [$C_{14}H_{22}(COOMe)_4$; m/e 426 (M^+); ν_{max}(CCl_4) 1740 cm^{-1}; δ_{ppm}(CCl_4) 0.88(3H, d, J=6.5Hz), 0.88(3H, t, J=6.2Hz), 3.57(3H, s), 3.59(6H, s), and 3.64(3H, s)]. Hydrolysis of the residue with 2N H_2SO_4 (reflux temp., 4 hr) gave L-ornithine and oxalic acid.

Thus, the relative positions among two amido groups and a carbonyl group have been unambiguously established. Therefore, the partial structure of ikarugamycin (1) is probably represented by I, for the formation of 5 indicates that two of the three -CH=CH- groups in 1 must be located as shown in I except for the geometries of the two conjugated double bonds.

In order to confirm the partial structure (I), the following reactions were carried out. Oxidation of 3 with CrO_3 in 6N H_2SO_4 (80°, 2 hr) gave a keto acid 6 [mp 222-223°;



1(I)



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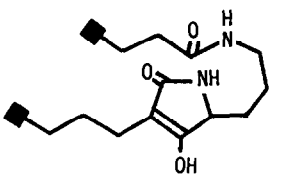

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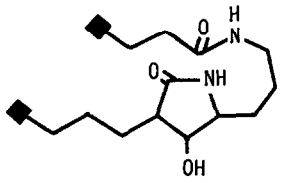

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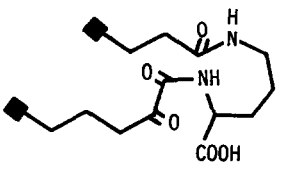

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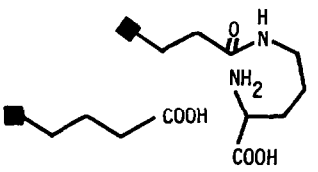

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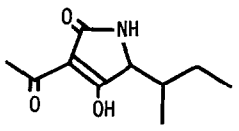
$C_{29}H_{46}O_5N_2 \cdot MeOH$], which was oxidized again with alkaline H_2O_2 to afford an amino acid 7 [m/e 562 (M^+); $\delta_{ppm}(CDCl_3)$ 2.04(3H, s), 3.68(3H, s), 3.75(3H, s), and 4.5(1H, m) as the N-acetyl dimethyl ester]. Hydrolysis of a DNP-derivative of the amino acid (7) with AcOH-6N HCl yielded α-DNP-ornithine and a tricyclic dicarboxylic acid which was isolated as the corresponding dimethyl ester 8 [$C_{21}H_{36}(COOMe)_2$; m/e 406 (M^+)]. This indicates that the partial structure I only is the possible one.

In fact, there are close similarities of the IR and UV spectra between hexahydroikarugamycin (2) and tenuazoic acid (9)[6] [$\nu_{max}(CHCl_3)$ 1710, 1660, and 1616 cm^{-1}; λ_{max}(EtOH) 217 and 277 nm (ε 5,100 and 12,900, respectively), λ_{max}(0.1N NaOH) 239 and 279 nm (ε 9,600 and 12,000, respectively)].



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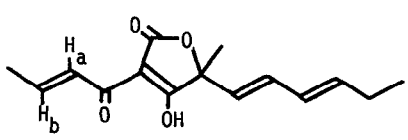

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In the NMR spectrum of 1, the signals of H_a (δ 6.96) and H_b (δ 6.64) protons, which are observed in the considerably lower field than those of the other four olefinic protons (δ 5.6~6.2), must be attached to the double bond conjugated with the tricarbonyl system.

In addition, it should be noted that the signal of H_a proton appears in the lower field than that of H_b proton. Such a case has been known in the NMR spectrum of aspertetronin A (10)[7] [δ 7.32 (H_a) and 7.06 (H_b)]. Accordingly, the geometries of the two double bonds in 1 can be assigned as shown in I.

Full structure of ikarugamycin (1) will be reported in the following paper.

Acknowledgments

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REFERENCES AND FOOTNOTES

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2. Satisfactory micro analytical results were obtained for all crystalline compounds.
3. Crystalline form and melting point (dec) of ikarugamycin varied with recrystallization solvent.
4. All asterisk-signals disappeared on addition of D_2O.
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