CHEMISTRY LETTERS, pp. 789-792, 1973. Published by the Chemical Society of Japan

# ABSOLUTE CONFIGURATIONS OF TWO SUGARS OF ANTIBIOTIC B-58941

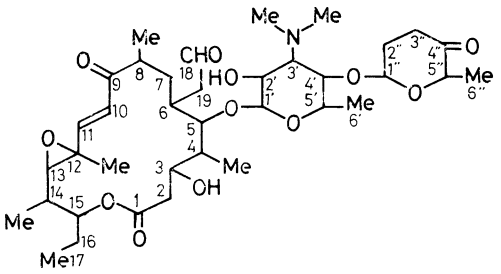
Takashi SUZUKI, Norio SUGITA, and Mitsuko ASAI

Microbiological Research Laboratories, Central Research Division,

Takeda Chemical Industries, Ltd., Higashiyodogawa-ku, Osaka 532

Absolute configurations of neutral sugar of antibiotic B-58941 and two methyl glycosides of its dihydro-sugar are assumed as 2,3,6-trideoxy-L-hexopyranos-4-ulose and methyl 2,3,6-trideoxy-(α or β)-L-erythro-hexopyranoside, respectively. Methyl glycoside of a basic sugar of B-58941-B is identified as methyl D-mycaminoside.

In the previous paper, the plain structure of an antibiotic B-58941[1] (I), $C_{37}H_{59}O_{12}N$, has been proposed to be I[1] on the basis of the NMR spectrum and spin-decoupling of its diacetate. When I was hydrolyzed with dilute hydrochloric acid for three hours at room temperature, a basic substance B-58941-B[1] (II), $C_{31}H_{51}O_{10}N$, was obtained from the lipophilic fraction and a neutral sugar[1] (III), $C_6H_{10}O_3$, was obtained from the hydrophilic fraction. Oxidation of III with sodium periodate gave one mole of acetaldehyde (2,4-DNPH, mp 164°C) and one mole of succinaldehydic acid which was identified with an authentic sample (2,4-DNPH, mp 202°C), and hence, it was assumed that III is 2,3,6-trideoxy-hexopyranos-4-ulose. Celmer has reported that this 4-keto sugar is an important biosynthetic intermediate glimpsed how the L-series can arise from the D-series.[2] In this paper, the absolute configurations of the sugars of the antibiotic are reported.



I

Treatment of I with sodium borohydride gave its reduction product, hexa-

hydro B-58941 (VI), $C_{37}H_{65}O_{12}N$, [mp 128°C, $[\alpha]_D^{22}$ -30.3° ($CHCl_3$), $\lambda_{max}^{MeOH}$ 207 nm ($\varepsilon$ = 9010), $\nu_{max}^{KBr}$ 3430 (OH), 1730 (lactone CO), 1630 (>C = C<), 1060 $cm^{-1}$ (-O-), $M^+$ 715], and VI was further acetylated with acetic anhydride in pyridine to give its pentaacetate (VII), $C_{47}H_{75}O_{17}N$, [mp 107°C, $\nu_{max}^{CHCl_3}$ 1740, 1245 $cm^{-1}$ ($-OCOCH_3$), NMR ($CDCl_3$, 100 MHz) $\delta$ 2.01-2.04 ppm ($5-COC\underline{H}_3$), $M^+$ 925]. When VI was treated with hydrochloric acid in methanol at 5°C for 16 hours, followed by adjustment of the reaction solution to pH 5.0, evaporation of methanol, addition of water and then extraction with chloroform, about one mole of methyl glycoside (IV) was obtained from the extract, and about one mole of tetrahydro B-58941-B (VIII), $C_{31}H_{55}O_{10}N$, [mp 123-125°C, $[\alpha]_D^{22}$ -3° ($CHCl_3$), $\nu_{max}^{KBr}$ 3420 (OH), 1725 (lactone CO), 1625 $cm^{-1}$ (>C = C<)] was obtained from the chloroform extract of the resulted aqueous layer adjusted to pH 8.5. IV was separated into two isomers by chromatography on silica gel; the main component being IVa, $C_7H_{14}O_3$, [colorless syrup, $[\alpha]_D^{22}$ -144.2° ($CHCl_3$), $\nu_{max}^{liq}$ 3400 (OH), 1160, 1130 $cm^{-1}$ (-O-), NMR ($CDCl_3$) $\delta$ 4.57 (t, J <3 Hz, anomeric proton), 3.94 (m, $-O-C\underline{H}-$), 3.53 (dq, J=9 and 6 Hz, $-O-C\underline{H}-CH_3$), 3.31 (s, $-OC\underline{H}_3$), 3.2 ($-O\underline{H}$), 1.8 (m, $2-C\underline{H}_2-$), 1.26 ppm (d, J=6 Hz, $-CH-C\underline{H}_3$)] and the minor component being IVb, $C_7H_{14}O_3$, [colorless syrup, $[\alpha]_D^{22}$ +27.2° ($CHCl_3$), $\nu_{max}^{liq}$ 3400 (OH), 1160, 1130 $cm^{-1}$ (-O-), NMR ($CDCl_3$), $\delta$ 4.30 (dd, J=9 and 3 Hz, anomeric proton), 3.84 (m, $-O-C\underline{H}-$), 3.52 (dq, J=9 and 6 Hz, $-O-C\underline{H}-CH_3$), 3.44 (s, $-OC\underline{H}_3$), 3.2 ($-O\underline{H}$), 1.6 (m, $2-C\underline{H}_2-$), 1.28 ppm (d, J=6 Hz, $-CH-C\underline{H}_3$)]. Since anomeric protons (H-1) of IVa and IVb are assigned at 4.57 ppm (t, $J_{1e,\,2a}$ <3 Hz, $J_{1e,\,2e}$ <3 Hz) and 4.30 ppm (dd, J=9 and 3 Hz), respectively, the conformation of H-1 of IVa and that of IVb are assumed to be equatorial and axial, respectively. When methyl protons (1.26 ppm, d, J=6 Hz) of IVa are irradiated, signal of H-5 (3.53 ppm, dq, J=9 and 6 Hz) is decoupled into a doublet (J=9 Hz), and when methylene protons (1.8 ppm, m) thereof are irradiated, signal of H-4 (3.94 ppm, m) is decoupled into a doublet (J=9 Hz), and hence, the conformational relation between H-4 and H-5 of IVa is concluded to be diaxial. That of H-4 and H-5 of IVb is also concluded to be diaxial from the same results of measurement as above.[3,4]

Therefore, IVa and IVb are anomers with each other, and their plain structure is methyl 2,3,6-trideoxy-hexopyranoside. On a reasonable assumption that both IVa and IVb have the chair conformation and a fact that the H-4 and H-5 protons lie in a diaxial relationship, only four structures (as shown below Fig. 1) may be possible for those of IVa and IVb, among the probable sixteen structures.[5] Since the conformation of H-1 of IVa is equatorial and that of IVb is axial,

the stereoisomers of IVa and IVb are confined to either α-D (C1) (A) or α-L (1C) (C) and either β-D (C1) (B) or β-L (1C) (D), respectively.[4] However, according to the Hudson's rule,[6] the more levorotary member of an α,β-pair of anomers is to be named α-L in L-series and β-D in D-series, IVa which is the more levorotary anomer must be either β-D or α-L. Consequently the absolute configuration of IVa is established as α-L (1C) (C), that is, methyl 2,3,6-trideoxy-α-L-erythro-hexopyranoside (1C). Likewise, the absolute configurations of the anomer IVb is established as β-L (1C) (D), that is, methyl 2,3,6-trideoxy-β-L-erythro-hexopyranoside (1C).

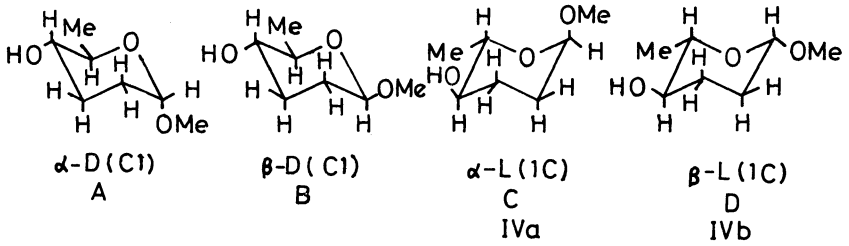
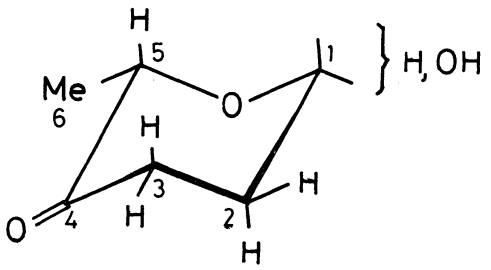



Fig.1. The possible structures of IVa and IVb

Comparing J-value of the H-1" (4.80 ppm, t, $J_{1e,\,2a}<3$ Hz, $J_{1e,\,2e}<3$ Hz) of VI with those of the H-1 of IVa and IVb, the linkage of 2,3,6-trideoxy-L-erythro-hexopyranose (IV) in VI is assumed to be α-linkage. If Klyne's rule[7] can be applied to these compounds, the linkage of IV in VI is assumed to be α-linkage, since $[M]_D$ of VI (-216°) is nearly equal to the sum of $[M]_D$ of VIII (-18°) and $[M]_D$ of IVa (-222°).[7] Further, it seems that by the reduction of I with sodium borohydride carbonyl group at C-4" of III was stereoselectively reduced to give only the equatorial C-4"-OH, and on the basis of the above, III is assumed to be 2,3,6-trideoxy-L-hexopyranos-4-ulose.




III

One of the methyl glycosides of the dihydro-III, methyl 2,3,6-trideoxy-α-L-erythro-hexopyranoside (IVa), seems to be identical with the methyl L-amicetoside[8] obtained by Keller-Schierlein and therefore, it is presumed that the neutral sugar (III) of B-58941 is identical with cinerulose[8] discovered in cinerubine A later on.

Secondary, when II was hydrolyzed by refluxing with 2 N HCl for three hours, followed by removal of insoluble substance, evaporation of the filtrate to dryness, about one mole of a basic sugar (V), $C_8H_{17}O_4N \cdot HCl$, [mp 115°C, $[\alpha]_D^{22}$ +28° ($H_2O$)] was obtained from the residue. When V was treated with hydrochloric acid in methanol at room temperature for three days, methyl glycoside (IX) was obtained. IX was separated into two isomers by chromatography on silica gel; IXa, $C_9H_{19}O_4N$, [mp 124°C, $[\alpha]_D^{22}$ -38° ($H_2O$), NMR ($CDCl_3$) $\delta$ 4.12 ppm (d, J=9 Hz, -O-C$\underline{H}$-)] and IXb, $C_9H_{19}O_4N$, [mp 84°C, $[\alpha]_D^{22}$ +112° ($H_2O$), NMR ($CDCl_3$) $\delta$ 4.60 ppm (d, J=4 Hz, -O-C$\underline{H}$-)]. These properties of IXa and IXb are identical with those of methyl β-D-mycaminoside and methyl α-D-mycaminoside,[9,10] respectively, which were obtained from carbomycin,[11] and hence, V is identified as D-mycaminose.[9]

## References


1) T. Suzuki, Bull. Chem. Soc. Japan, 43, 292 (1970).

2) W. D. Celmer, Pure and Applied Chemistry, 28, 413 (1971).

3) M. Karplus, J. Chem. Phys. 30, 11 (1959); idem., J. Am. Chem. Soc., 85, 2870 (1963).

4) Lemieux, R. U., R. K. Kulling, H. J. Bernstein and W. G. Schneider, J. Am. Chem. Soc., 79, 1005 (1957).

5) Reeves, R. E., J. Am. Chem. Soc., 72, 1499 (1950).

6) G. S. Hudson, J. Am. Chem. Soc., 31, 66 (1909).

7) W. Klyne, Biochem. J., 47, xli (1950).

8) W. Keller-Schierlein and W. Richle, Antimicrobial Agents and Chemotherapy - 1970, 68 (1971).

9) F. A. Hochstein and P. P. Regna, J. Am. Chem. Soc., 77, 3353 (1955).

10) S. Omura, M. Katagiri, H. Ogura and T. Hata, Chem. Pharm. Bull. (Tokyo), 16, 1167 (1968).

11) F. W. Tanner, Jr., A. R. English, T. M. Lees and J. B. Routin, Antibiotics & Chemotherapy, 2, 441 (1952).


( Received March 26, 1973 )