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Structural Studies of Fortimicins. II.† Crystal and Molecular Structure of Fortamine

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Fortamine, L-3-amino-1-methoxy-6-methylamino-1,3,6-trideoxy-*chiro*-inositol, is a component of fortimicins which are potent aminocyclitol antibiotics. The space group is $P2_1$ with $a=9.454(1)$, $b=6.6957(8)$, $c=7.622(1)$ Å, $\beta=95.93(2)°$, and two $C_8H_{18}N_2O_4$'s per unit cell. The structure was solved by direct methods, least-squares refinement using 932 reflexions giving the final R value of 0.033. The ring conformation of fortamine is closer to the ideal chair form than that of the fortamine moiety in fortimicin B. The strain caused by the intramolecular hydrogen bond and glycosidic bond in fortimicin B is released in fortamine.

Fortimicins[1,2] are potent broad-spectrum aminocyclitol antibiotics produced by a strain of *Micromonospora olivoasterospona*.[3–5] The conformation of the 1,4-diaminocyclitol moiety of fortimicins is probably related to their activity.[1,3,4] We have determined the absolute configuration of fortimicin B (Fig. 1) by X-ray analysis.[6] An intramolecular hydrogen bond is formed between O(5)–H and N(2′), the fortamine ring being distorted from the ideal chair conformation. It is of interest to examine whether the ring distortion is due to the glycosidic bond or the intramolecular hydrogen bond. We have carried out an X-ray analysis of fortamine obtained by the hydrolysis of fortimicin B.

Experimental and Structure Determination

Fortamine was prepared by the hydrolysis of fortimicin B with hydrochloric acid. Crystals of fortamine were grown from a methanol solution. A $0.5\times0.3\times0.3$ mm^3 crystal sealed in a glass capillary was used for data collection on a Rigaku four-circle automated diffractometer with graphite-monochromated Mo $K\alpha$ radiation ($\lambda=0.71069$ Å). Preliminary unit cell dimensions and space group were obtained from photographs. The space group was determined from systematic absence ($0k0$ for odd k). Accurate cell dimensions were determined by least-squares calculations with the 2θ values of 15 high-angle reflexions measured on the diffractometer. Crystal data are summarized in Table 1. All reflexions within the range $2\theta\leqq55°$ were collected by use of the ω-2θ scan mode with a scanning rate of $4°(2\theta)$ min^{-1}. Stationary background counts were accumulated for 10 s before and after each scan. Periodic checks of the intensity values of three standard reflexions showed no significant X-ray damage or crystal decay. Corrections for absorption or extinction were not applied. A total of 1191 independent reflexions were obtained, of which 932 ($|F_0|>3.0\sigma(|F_0|)$) were considered as observed.

The phases of 185 reflexions with $|E_0|\geqq1.2$ were assigned with MULTAN.[7] The best set of phases was used to calculate an E map, which gave 14 chemically significant peaks. The structural parameters were refined by block-diagonal least-squares methods with a modified HBLS program. At the stage $R=0.17$, all the hydrogen atoms were found by the difference Fourier synthesis. Refinement using anisotropic and isotropic temperature factors for the non-hydrogen and hydrogen atoms, respectively, gave the final R value 0.033 for 932 reflexions. The weighting system used in the final stage was $w=0.3$ for $|F_0|<1.51$ and $|F_0|>35.57$, and $w=(0.00792(F_0)^2-0.29822|F_0|+3.76458)^{-1}$ for $1.51\leqq|F_0|\leqq35.57$. Six strong reflexions, 1 −1 0, 1 −4 1, −1 −1 4, 1 0 0, −2 3 3, and −3 4 3, were excluded because they seemed to suffer from secondary extinctions. Atomic scattering factors were taken from "International Tables for X-Ray Crystallography".[8] The final positional and thermal parameters are given in Table 2. The tables of observed and calculated structure factors are kept as Document No. 8024 at the Chemical Society of Japan.

TABLE 1. CRYSTAL DATA

$C_8H_{18}N_2O_4$	$b=6.6957(8)$
$M.W.=206.24$	$c=7.622(1)$ Å
$P2_1$	$\beta=95.93(2)°$
$Z=2$	$D_x=1.4276$ g·cm^{-3}
$a=9.454(1)$	μ(Mo $K\alpha$)$=1.221$ cm^{-1}

Results and Discussion

Hydrogen Bond and Crystal Structure. The numbering system of atoms and the crystal structure viewed along the b axis are shown in Figs. 1 and 2, respectively. Hydrogen bond lengths and angles are given in Table 3. In the fortamine moiety of fortimicin B, only the O(2) hydroxyl group is involved in the intermolecular hydrogen bond. In fortamine, on the other hand, all functional groups except the methoxyl are involved in the intermolecular hydrogen bonds, there being no

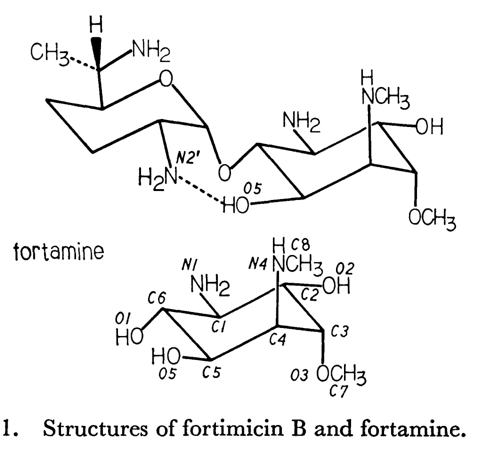

Fig. 1. Structures of fortimicin B and fortamine.

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† Part I of the series: "Structure of Fortimicin B."[6]

Table 2a. Final atomic coordinates with their estimated standard deviations, multiplied by 10^4 for non-hydrogen atoms and 10^3 for hydrogen atoms

Atom	x	y	z	Atom	x	y	z
C(1)	967(2)	3690(4)	2962(3)	HC(3)	368(2)	125(4)	135(3)
C(2)	1780(3)	1930(4)	2320(3)	HC(4)	420(2)	464(4)	35(3)
C(3)	3247(2)	2514(4)	1869(3)	HC(5)	301(3)	640(4)	236(3)
C(4)	3162(2)	4201(4)	502(3)	HC(6)	26(2)	500(4)	51(3)
C(5)	2372(3)	5985(4)	1179(3)	HAC(7)	528(3)	96(6)	407(4)
C(6)	897(3)	5422(4)	1642(3)	HBC(7)	602(3)	334(6)	475(4)
C(7)	5428(3)	2351(5)	3765(4)	HCC(7)	593(3)	249(6)	280(4)
C(8)	3352(3)	2146(5)	−2127(3)	HAC(8)	293(2)	207(4)	−336(3)
N(1)	−488(2)	3057(4)	3223(3)	HBC(8)	329(3)	75(5)	−149(4)
N(4)	2458(2)	3532(3)	−1229(2)	HCC(8)	434(2)	261(4)	−211(3)
O(1)	266(2)	7113(3)	2411(2)	HAN(1)	−100(3)	426(5)	367(3)
O(2)	1874(2)	399(3)	3640(2)	HBN(1)	−45(3)	209(5)	400(3)
O(3)	4074(2)	3243(3)	3421(2)	HN(4)	230(2)	467(4)	−192(3)
O(5)	2347(2)	7648(3)	1(2)	HO(1)	−50(2)	737(4)	189(3)
HC(1)	141(2)	418(4)	402(3)	HO(2)	144(3)	−59(4)	328(3)
HC(2)	127(2)	137(4)	122(3)	HO(5)	167(3)	751(5)	−92(3)

Table 2b. Final thermal parameters with their estimated standard deviations
Anisotropic thermal parameters (Å^2) are multiplied by 10^4 and isotropic ones (Å^2) multiplied by 10^3.

Atom	U or U_{11}	U_{22}	U_{33}	U_{12}	U_{13}	U_{23}
C(1)	284(1)	241(13)	226(11)	−30(11)	−7(9)	−13(11)
C(2)	371(13)	194(12)	197(11)	−4(12)	−40(10)	26(10)
C(3)	320(12)	280(15)	219(11)	50(12)	−36(9)	−74(11)
C(4)	215(11)	314(15)	276(12)	−43(11)	29(10)	−53(12)
C(5)	298(13)	235(13)	260(12)	−20(11)	−10(10)	−21(11)
C(6)	265(12)	213(12)	242(11)	34(11)	−3(9)	−35(11)
C(7)	302(14)	508(19)	488(16)	53(15)	−71(12)	12(16)
C(8)	433(15)	429(17)	275(13)	39(15)	67(11)	−59(14)
N(1)	343(11)	321(13)	330(11)	−7(11)	88(9)	39(11)
N(4)	275(10)	273(12)	224(9)	−1(10)	24(8)	5(10)
O(1)	286(8)	245(9)	386(9)	66(9)	−16(7)	−68(9)
O(2)	518(11)	215(9)	297(9)	−6(9)	−46(8)	55(9)
O(3)	290(9)	458(12)	336(9)	115(10)	−112(7)	−120(10)
O(5)	429(10)	257(10)	389(10)	−61(9)	13(8)	68(9)
HC(1)	23(6)					
HC(2)	25(6)					
HC(3)	24(7)					
HC(4)	22(6)					
HC(5)	44(8)					
HC(6)	21(6)					
HAC(7)	87(12)					
HBC(7)	78(11)					
HCC(7)	96(12)					
HAC(8)	38(7)					
HBC(8)	63(9)					
HCC(8)	43(8)					
HAN(1)	60(9)					
HBN(1)	53(8)					
HN(4)	27(7)					
HO(1)	51(8)					
HO(2)	43(8)					
HO(5)	58(9)					

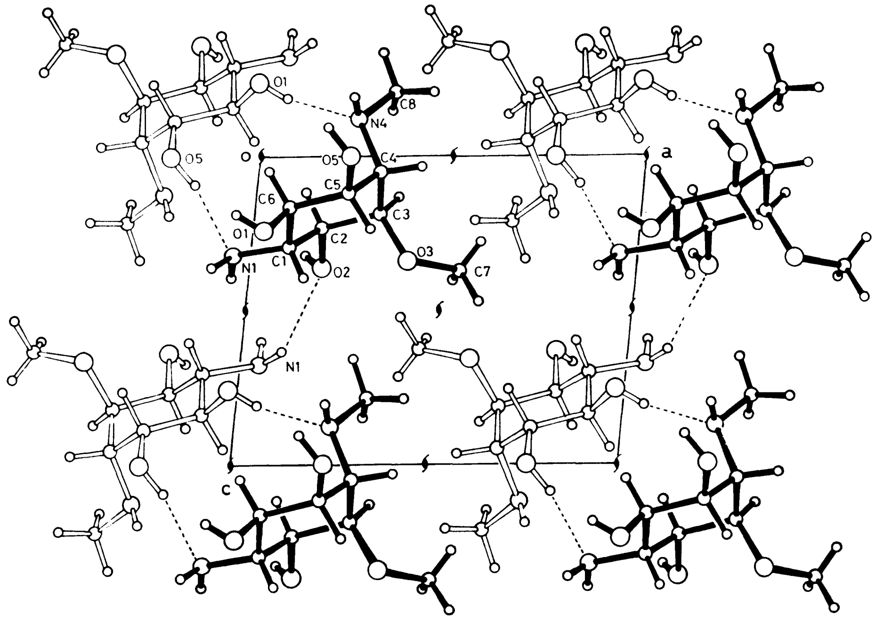

Fig. 2. The crystal structure projected along the b axis. The molecule composed of the numbering atoms corresponds to that given in Table 2.

TABLE 3. HYDROGEN BOND DISTANCES (l/Å) AND ANGLES (ϕ/°)

A–H···B	A···B	∠A–H···B
O(1)–H^{I}···N(4)^{II}	2.806 (3)	163 (3)
O(5)–H^{I}···N(1)^{II}	2.881 (3)	160 (3)
O(2)–H^{I}···O(1)^{III}	2.781 (3)	177 (3)
N(1)–H^{I}···O(2)^{IV}	3.246 (3)	138 (3)

I: $(x\ y\ z)$
II: $(-x\ 1/2+y\ -z)$
III: $(x\ -1+y\ z)$
IV: $(-x\ 1/2+y\ 1-z)$

intramolecular hydrogen bond. The molecules related by the 2_1 screw axis through the origin are connected by the O(1)–H···N(4) and O(5)–H···N(1) hydrogen bonds. Along the b axis the molecules are linked by the O(2)–H···O(1) hydrogen bonds. The infinite columns thus formed are further interlinked by the weak N(1)–H···O(2) hydrogen bond to give double-layers. Boundary between the double-layers at $(1/2, y, z)$ is held together mainly by the van der Waals forces of *N*-methyl and *O*-methyl groups.

Bond Lengths and Angles. Bond lengths and angles are given in Table 4, together with the corresponding values in fortimicin B. The average endocyclic

TABLE 4. BOND LENGTHS (l/Å) AND ANGLES (ϕ/°)

	Fortamine	Fortimicin B		Fortamine	Fortimicin B
C(1)–C(2)	1.505(4)	1.524(8)	C(1)–C(2)–C(3)	112.6(2)	110.4(5)
C(1)–C(6)	1.545(4)	1.510(8)	C(2)–C(3)–C(4)	111.1(2)	111.3(5)
C(2)–C(3)	1.513(4)	1.523(8)	C(3)–C(4)–C(5)	109.9(2)	109.3(5)
C(3)–C(4)	1.532(4)	1.526(9)	C(4)–C(5)–C(6)	112.0(2)	113.5(5)
C(4)–C(5)	1.528(4)	1.546(9)	C(5)–C(6)–C(1)	111.2(2)	113.9(5)
C(5)–C(6)	1.521(4)	1.501(8)	C(6)–C(1)–C(2)	111.4(2)	109.6(5)
C(1)–N(1)	1.470(4)	1.474(8)	N(1)–C(1)–C(2)	110.4(2)	110.2(5)
C(2)–O(2)	1.432(3)	1.410(7)	N(1)–C(1)–C(6)	108.7(2)	108.2(5)
C(3)–O(3)	1.434(4)	1.427(7)	O(2)–C(2)–C(1)	108.7(2)	111.1(5)
O(3)–C(7)	1.414(4)	1.389(10)	O(2)–C(2)–C(3)	110.7(2)	112.3(5)
C(4)–N(4)	1.485(4)	1.455(9)	O(3)–C(3)–C(2)	109.6(2)	113.6(5)
N(4)–C(8)	1.469(4)	1.486(9)	O(3)–C(3)–C(4)	107.2(2)	105.2(5)
C(5)–O(5)	1.429(3)	1.425(7)	C(7)–O(3)–C(3)	114.5(2)	116.6(5)
C(6)–O(1)	1.434(3)	1.439(7)	C(8)–N(4)–C(4)	112.2(2)	113.4(5)
			N(4)–C(4)–C(3)	111.8(2)	112.6(5)
			N(4)–C(4)–C(5)	110.3(2)	109.1(5)
			O(5)–C(5)–C(4)	112.0(2)	107.5(5)
			O(5)–C(5)–C(6)	112.5(2)	112.8(5)
			O(1)–C(6)–C(5)	109.1(2)	107.4(5)
			O(1)–C(6)–C(1)	109.1(2)	108.5(5)

C–C bond length, 1.522 Å, is equal to that of fortimicin B. The value is essentially the same as that of cyclohexane (1.532 Å)[9] and *myo*-inositol (1.521 Å).[10] The bond shortening of C(1)–C(6) and C(5)–C(6) in fortimicin B might be due to the glycosidic linkage. The long C(4)–C(5) distance of 1.546 Å in fortimicin B is probably caused by the strong intramolecular hydrogen bond. The results and geometries of cyclohexane rings in other compounds[9–13] suggest that the endocyclic C–C bond lengths can easily vary from 1.50 to 1.55 Å according to the intra- and intermolecular interactions.

The C(2)–O(2) and C(4)–N(4) bond lengths in fortamine are greater than those in fortimicin B by over 3σ. While O(2) in fortimicin B is weakly hydrogen bonded to solvent molecules with an O···O distance of 2.926(7) Å, that in fortamine is fairly strongly hydrogen bonded to the O(1) hydroxyl group with an O···O distance of 2.781(3) Å. Similarly, the N(4) atom in fortimicin B does not participate in a hydrogen bond, but that in fortamine is hydrogen bonded to the O(1) hydroxyl group. Such hydrogen bonds, therefore, seem to lengthen the bonds of C(2)–O(2) and C(4)–N(4) in fortamine.

The endocyclic bond angles C(1)–C(6)–C(5) and C(4)–C(5)–C(6) in fortimicin B are 113.9 and 113.5°, respectively, significantly larger than the average angle 111.5° in cyclohexane.[9] The corresponding values in fortamine having neither a glycosidic linkage nor an intramolecular hydrogen bond are 111.2 and 112.0°. The relatively large C(1)–C(2)–C(3) angle of 112.6° is probably due to the strong O(2)–H···O(1) hydrogen bond. The exocyclic bond angles of C(4)–C(5)–O(5) and O(5)–C(5)–C(6) are asymmetrical in fortimicin B because of the intramolecular hydrogen bond, while in fortamine they are symmetrical. Although O(3)–C(3)–C(4) and O(3)–C(3)–C(2) angles in fortimicin B are asymmetrical, they are symmetrical in fortamine.

Bond lengths and angles in fortamine are closer to the normal values than those in fortimicin B. Thus the glycosidic linkage and the intramolecular hydrogen bond might influence the local bond lengths and angles of fortamine ring.

TABLE 5. TORSIONAL ANGLES (τ/°)

	Fortamine	Fortimicin B
C(6)–C(1)–C(2)–C(3)	53.9	58.0
C(1)–C(2)–C(3)–C(4)	−56.1	−61.0
C(2)–C(3)–C(4)–C(5)	56.3	55.6
C(3)–C(4)–C(5)–C(6)	−56.4	−50.4
C(4)–C(5)–C(6)–C(1)	54.6	50.9
C(5)–C(6)–C(1)–C(2)	−52.6	−53.6
N(1)–C(1)–C(2)–O(2)	−62.2	−57.7
O(2)–C(2)–C(3)–O(3)	−59.8	−67.1
C(7)–O(3)–C(3)–C(4)	−110.1	−152.7
C(7)–O(3)–C(3)–C(2)	129.2	85.4
O(3)–C(3)–C(4)–N(4)	173.9	170.7
C(8)–N(4)–C(4)–C(5)	166.5	167.0
C(8)–N(4)–C(4)–C(3)	−70.9	−71.5
N(4)–C(4)–C(5)–O(5)	−60.1	−52.3
O(5)–C(5)–C(6)–O(1)	−57.9	−66.4
O(1)–C(6)–C(1)–N(1)	65.2	66.7

Conformation of the Molecule. A stereoscopic drawing of the molecule is shown in Fig. 3. The conformations of the ring and substituents of fortamine are essentially the same as those of fortimicin B. All torsional angles in the molecule are compared in Table 5. The overall flattening of fortamine rings is essentially the same in these two compounds with mean C–C–C–C torsional angles of 54.9° for fortimicin B and 55.0° for fortamine. These values are close to 54.9° in cyclohexane as determined by electron diffraction study.[9] In *epi*- and *myo*-inositol,[10] however, the ring is more puckered with the mean C–C–C–C torsional angles of 56.3 and 56.8°, respectively.

Closer examination shows that the present molecule takes a more symmetrical conformation than fortimicin B. The characteristics of the ring conformation are illustrated in Fig. 4, where the distance, d, is defined as the deviation of the two atoms from the least-squares plane of the other four atoms forming the seat of a chair. They are good indicies to show the puckering or the flattening of cyclohexane ring. The flattening, d= 0.601 Å, of the C(4)–C(5)–C(6) part in fortimicin B by the intramolecular hydrogen bond is accompanied

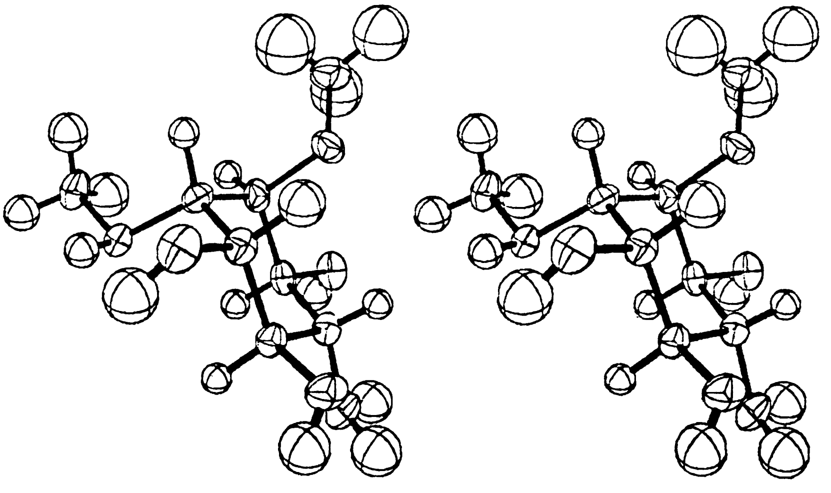

Fig. 3. Stereoscopic view with thermal ellipsoids at 50% probability.

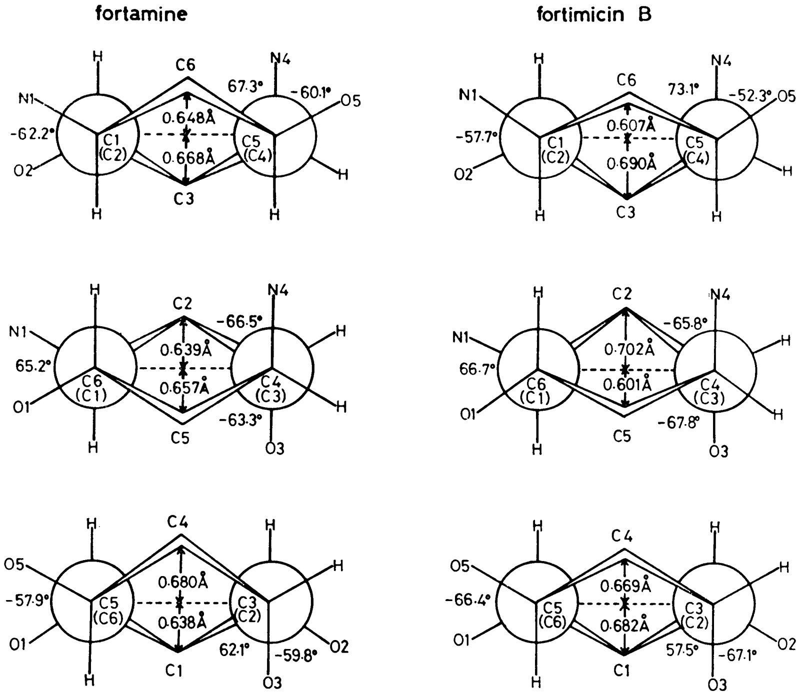

Fig. 4. Schematic drawing of chair forms of fortamine and fortimicin B.

by the puckering of the C(1)–C(2)–C(3) part, $d=0.702$ Å. The C(1)–C(6)–C(5) part in fortimicin B is flattened, ($d=0.607$ Å), probably due to the effect of the glycosidic bond, the flattening being followed by the puckering of the C(2)–C(3)–C(4) part, $d=0.690$ Å. In fortamine a slight puckering, $d=0.680$ Å, is seen at the *N*-methyl part but the ring as a whole takes essentially a symmetric structure.

These characteristics are related to the torsional angles. The C(3)–C(4)–C(5)–C(6), C(4)–C(5)–C(6)–C(1), C(1)–C(2)–C(3)–C(4) and C(6)–C(1)–C(2)–C(3) angles are significantly different between fortamine and fortimicin B. It seems that the remarkably small values of C(3)–C(4)–C(5)–C(6) and C(4)–C(5)–C(6)–C(1) angles in fortimicin B are mainly due to the intramolecular hydrogen bond. The effect of this local deformation of the chair geometry of the ring is discussed by calculating minimum energy conformation.[14] The calculation shows that the deformation of a torsional angle around an endocyclic bond induces an inverse deformation of the torsional angle around the opposite endocyclic bond in the ring. The extent is about 30% of the original deformation. The increase in C(1)–C(2)–C(3)–C(4) and C(6)–C(1)–C(2)–C(3) angles in fortimicin B are explained by this effect.

The results indicate that the glycosidic bond and the intramolecular hydrogen bond considerably flatten the fortamine ring locally.

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