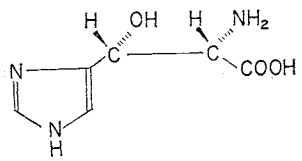
Communications to the editor

THE CHEMISTRY OF BLEOMYCIN. X*

THE STEREOCHEMISTRY AND CRYSTAL STRUCTURE OF β-HYDROXYHISTIDINE, AN AMINE COMPONENT OF BLEOMYCIN

Sir:

On acid hydrolysis of bleomycin A_2, one of the diastereoisomers of β-hydroxyhistidine was obtained[1]. In order to establish the stereochemistry, X-ray analysis of the monohydrobromide was undertaken. In the present paper, the stereochemistry is shown to be L-*erthro*-β-hydroxyhistidine (**I**).



I

Crystals of the hydrobromide of compound I were prepared by recrystallization from aqueous alcohol. The crystal data are as follows:

Space group:
Monoclinic, $P2_1$

Systematic absences:
$0k0$, $k=2n+1$

$a=10.35_6$ Å
$b=6.08_6$ Å
$c=7.11_4$ Å
$\beta=96.4_6$
$V=455.5$ Å^3

Molecules per unit cell: 2 of $C_6H_9N_3O_3\cdot HBr$ (m.w. 252.06).

The lattice constants and three-dimensional intensity data were measured on a Rigaku Denki four-circle diffractometer up to $2\theta_{max.}=60°$ by the use of molybdenum radiation ($\lambda=0.71068$ Å). After LORENTZ and polarization corrections were applied, the absolute scale and the over-all temperature factors were obtained by a WILSON plot. No absorption correction was made. A total of 1111 structure factors was estimated. The structure was solved by the conventional heavy atom method. The position of the bromine atom was easily determined from a three-dimensional PATTERSON function. A bromine-phased electron density map was then calculated ($R=26.6$ %) and all of the non-hydrogen atoms which corresponded to a plausible structure were recognized in the map. A subsequent FOURIER calculation with all thirteen non-hydrogen atoms gave a R-factor of 14.4 % and all carbon, nitrogen and oxygen atoms could be assigned at this stage. By four cycles of full-matrix least squares refinement varying isotropic temperature factors the R-factor was reduced down to 6.85 %. Further four cycles refinement by the least squares calculation, assuming anisotropic temperature factors for all non-hydrogen atoms, enabled seven hydrogen atoms to be determined from the difference FOURIER map ($R=5.74$ %). The absolute configuration of the molecule was determined to be the L-series of amino acids with respect to the α-carbon atom (C(2)) by use of the

Fig. 1.

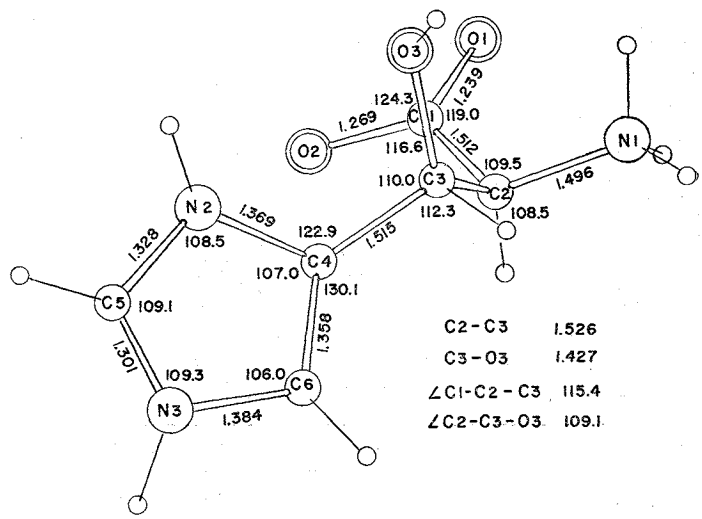


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Table 1. Final atomic parameters

Temperature factors are of the form $T=\exp[-(\beta_{11}h^2+\beta_{22}k^2+\beta_{33}l^2+2\beta_{12}hk+2\beta_{13}hl+2\beta_{23}kl)]$

	x	y	z	β_{11} or B	β_{22}	β_{33}	β_{12}	β_{13}	β_{23}
Br	0.0546(1)	0.0000(0)	0.3123(1)	0.0057(0)	0.0169(1)	0.0112(1)	0.0010(2)	−0.0003(1)	−0.0010(2)
O1	0.7810(5)	0.8240(8)	−0.0074(7)	0.0068(5)	0.0127(12)	0.0130(9)	0.0006(7)	0.0021(5)	0.0031(9)
O2	0.6122(4)	0.7412(9)	0.1454(7)	0.0055(5)	0.0198(15)	0.0134(9)	0.0027(7)	0.0025(5)	0.0014(10)
O3	0.7088(5)	0.2585(8)	0.0064(6)	0.0071(5)	0.0137(13)	0.0111(8)	−0.0010(7)	0.0021(5)	0.0004(9)
N1	0.9386(5)	0.5237(16)	0.1560(7)	0.0042(4)	0.0137(21)	0.0152(11)	−0.0011(9)	0.0010(5)	0.0033(14)
N2	0.5029(5)	0.2795(10)	0.2235(7)	0.0048(5)	0.0156(15)	0.0099(9)	−0.0010(7)	0.0009(5)	0.0004(11)
N3	0.4902(9)	0.2599(17)	0.5213(12)	0.0061(6)	0.0150(15)	0.0137(16)	−0.0011(8)	0.0028(7)	0.0014(13)
C1	0.7287(6)	0.7153(10)	0.1103(9)	0.0046(5)	0.0107(15)	0.0100(11)	0.0015(7)	−0.0006(6)	−0.0021(11)
C2	0.8057(6)	0.7357(10)	0.2187(8)	0.0042(4)	0.0088(22)	0.0102(9)	−0.0012(7)	0.0010(5)	0.0004(10)
C3	0.7449(6)	0.3072(12)	0.2015(9)	0.0044(5)	0.0157(18)	0.0100(11)	0.0008(8)	0.0009(6)	0.0011(12)
C4	0.6278(6)	0.2871(12)	0.3101(9)	0.0048(5)	0.0155(18)	0.0099(11)	−0.0002(8)	0.0012(6)	0.0008(12)
C5	0.4227(6)	0.2613(12)	0.3557(10)	0.0049(5)	0.0119(16)	0.0142(13)	−0.0014(8)	0.0019(7)	0.0011(13)
C6	0.6209(6)	0.2752(12)	0.4995(9)	0.0052(5)	0.0156(18)	0.0104(11)	−0.0009(9)	0.0010(6)	0.0019(13)
H(C2)	0.820(5)	0.569(9)	0.344(7)	2.0					
H(C3)	0.819(6)	0.200(12)	0.279(9)	1.6(1.4)					
H(C5)	0.327(8)	0.228(16)	0.311(11)	3.4(1.9)					
H(C6)	0.691(6)	0.256(11)	0.605(8)	2.0					
H(N11)	0.932(6)	0.542(16)	0.013(10)	2.4(1.7)					
H(N12)	0.973(6)	0.657(13)	0.181(9)	1.3(1.3)					
H(N13)	0.999(7)	0.433(12)	0.209(10)	2.3(1.6)					
H(N2)	0.470(6)	0.276(11)	0.099(8)	2.0					
H(N3)	0.462	0.270	0.660	2.0					
H(O3)	0.734	0.100	−0.038	2.0					

anomalous dispersion effect due to the bromine atoms. Five additional cycles of the least squares refinement using anisotropic temperature factors for the non-hydrogen atoms and isotropic ones for hydrogen atoms gave a *R*-factor of 3.99 %. Finally all ten hydrogen atoms could be recognized at reasonable positions which gave acceptable bond lengths and angles and hydrogen bonds. Final *R*-factor was 3.95 % for 1111 reflections. The final atomic parameters are given in Table 1.

Thus, the molecular structure of I is concluded to be L-*erthro*-*β*-hydroxyhistidine. The intramolecular bond distances and angles are shown in Fig. 1 and their average standard deviations are 0.009 Å and 0.58°. One of the outstanding features of the structure is the conformation around the C(2)−C(3) bond; the imidazole is *gauche* to the carboxyl group and *trans* to the amino group, while the hydroxyl group is *gauche* to the both groups. It is apparent that the introduction of a hydroxyl group on the *β*-carbon atom (C(3)) causes the imidazole to be in an unusual position in comparison with that found in published data for various histidines**. The imidazole ring is positively charged and the five ring atoms and C(3) atom are nearly coplanar with a maximum displacement of 0.004 Å for the C(5) atom.

Gunji Koyama
Hikaru Nakamura
Yasuhiko Muraoka
Tomohisa Takita
Kenji Maeda
Hamao Umezawa

Institute of Microbial Chemistry
Kamiosaki, Shinagawa-ku,
Tokyo, Japan

** To date two types of the conformation have been reported for histidines; the folded conformation in which the imidazole ring is *gauche* to both the carboxyl and amino groups, was found in L-histidine hydrochloride monohydrate[2]; the extended conformation in which the imidazole ring is *trans* to the carboxyl and *gauche* to the amino group, was found in DL-histidine hydrochloride dihydrate[3], orthorhombic L-histidine[4], monoclinic L-histidine[5] and DL-histidine[6].

Yoichi Iitaka
University of Tokyo,
Faculty of Pharmaceutical
Sciences
Bunkyo-ku, Tokyo, Japan

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