Communications to the editor

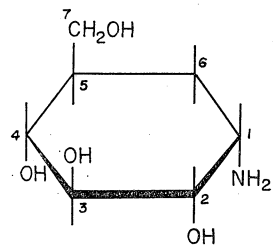
STUDIES ON VALIDAMYCINS, NEW ANTIBIOTICS. VII

THE X-RAY ANALYSIS OF VALIDAMINE HYDROBROMIDE

Sir:

The structure of validamine, **1**, has been determined by the X-ray analysis of its hydrobromide. In this communication we wish to report outlines of the structure elucidation.

Chart 1.



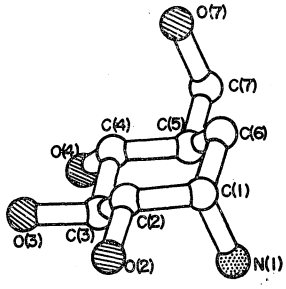
Validamycins[1] are new antibiotics which have been isolated from a culture filtrate of *Streptomyces hygroscopicus* var. *limoneus*. In connection with chemical degradation studies[2] on the structure of these antibiotics, X-ray analyses of their heavy-atom derivatives were undertaken. Most of them were very hygroscopic and it was difficult to get crystals suitable for the crystallographic use, but validamine hydrobromide ($C_7H_{15}NO_4 \cdot HBr$)[3], one of the degradation products obtained by hydrogenolysis and acid hydrolysis of validamycin A, was found to give promising crystals. We have accordingly undertaken the X-ray analysis of this compound.

The unit-cell, which contains four molecules of validamine hydrobromide, is orthorhombic with dimensions $a=10.44$, $b=14.21$ and $c=6.66$ Å. Systematic extinctions of hOO, OkO and OOl when h, k and l is odd, respectively, lead uniquely to the space group $P2_12_12_1$. Intensities of 802 reflections were measured on a HILGER and WATTS linear diffractometer with MoKα radiation. The coordinates of the bromine atom were determined from the three-dimensional sharpened PATTERSON function. For the detection of the lighter atoms, a three-dimensional minimum function and a FOURIER map based on the heavy-atom were calculated. By alternating application of least-squares refinement and FOURIER syntheses, all of the atomic positions (except hydrogen) were determined.

The identification of the various carbon, nitrogen and oxygen atoms was tested by the behavior of their temperature factors in the least-squares refinement. When all atoms except bromine were assumed as carbon, the isotropic temperature factors of three atoms attached to the six-membered ring at C(2), C(3) and C(4) become less than 1.0 Å^2. Accordingly, these atoms were assigned as oxygen (O(2), O(3) and O(4)). At the same time, temperature factors of the atom attached to the ring at C(1) and of the terminal atom bonded to C(7) became 1.6 and 2.6 Å^2, respectively. As these values were smaller than that of C(7) (2.9 Å^2), one of them should be identified as oxygen and the other as nitrogen. Since the temperature factors of five atoms directly attached to the cyclohexane ring should have nearly equal values, the atom bonded to C(1) was assigned as nitrogen (N(1)) and the other bonded to C(7) was assigned as oxygen (O(7)).

The refinement of atomic coordinates and temperature factors was carried out using the block-diagonal least-squares treatment with isotropic temperature factors. The discrepancy index decreased to 0.137. The isotropic thermal parameters of five atoms directly attached to the cyclohexane ring had nearly equal values; C(7): 2.89, O(2): 3.11, O(3): 3.36, O(4): 2.42 and N(1): 2.79 Å^2, respectively. These values seemed to be reasonable when compared with those of carbon atoms contained in the ring (2.1~2.7 Å^2). Relatively large standard deviations (0.03 Å) in their bond lengths, probably caused by the large dispersion effect of the bromine atom for MoKα radiation, left some uncertainties as to the identifications of these five atoms, so the assignments were reexamined by means of the studies on the nmr spectrum

Fig. 1. Projection of the structure seen down the *c* axis.



of the pentaacetate of validamine[3]. In Fig. 1 is given a projection of the structure down the *c* axis. Inter-atomic distances and angles for covalent bonds evidenced by the analysis are shown in Fig. 2. The molecule consists of a penta-substituted cyclohexane ring which is in a chair conformation. Three hydroxyl groups attached to C(2), C(3) and C(4) and a hydroxymethyl group attached to C(5) take an equatorial configuration, while an amino group attached to C(1) takes an axial configuration.

The absolute configuration of the molecule was also determined without ambiguity by the anomalous dispersion method, and the configuration represented in **1** (and in Fig. 1) was shown to be correct also in an absolute sense. Certain pairs of calculated structure factors $F(hkl)$ and $F(\bar{h}\bar{k}\bar{l})$ differed up to 50 % taking the dispersion corrections into account.

The structure of validamine was established from the present analysis to be 1S-(1,2,4/3,5)-1-amino-5-hydroxymethyl-2,3,4-cyclohexanetriol, **1**.

Acknowledgement

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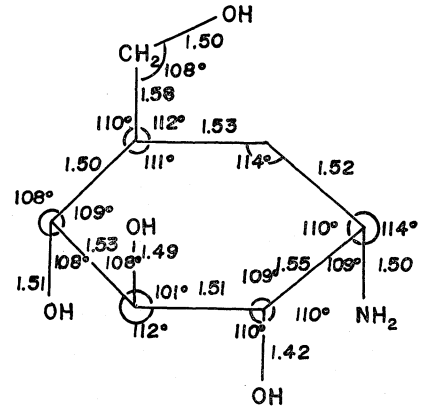
Fig. 2. Interatomic distances and angles.



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