Tetrahedron Letters No.34, pp. 3003-3006, 1970. Pergamon Press. Printed in Great Britain.

THE CRYSTAL AND MOLECULAR STRUCTURE OF 5-HYDROXY-DL-TRYPTOPHAN

Akio Wakahara, Masaru Kido, Takaji Fujiwara and Ken-ichi Tomita

Faculty of Pharmaceutical Sciences, Osaka University

Toyonaka, Osaka, Japan

(Received in Japan 30 April 1970; received in UK for publication 23 June 1970)

5-Hydroxytryptophan (5-HTP) is a metabolic precursor of serotonin, 5-hydroxytryptamine (5-HT), which is well known as one of the biogenic amines of pharmacodynamic action. 5-HT has been reported to have an excellent radiation protective ability[1]. Furthermore, Kobayashi et al.[2] have shown that 5-HTP might have a protective effect against lethal dose of ionizing radiation in mice.

It is of particular interest to determine the crystal and molecular structure of these radioprotective agents in order to elucidate the correlation between the stereostructure and its characteristic action.

In this paper, we report the crystal and molecular structure of 5-hydroxy-DL-tryptophan, and also compare its structure with other related structures, serotonin-creatinine sulphate complex[3], L-tryptophan hydrochloride[4] and 3-indolylacetic acid[5].

Crystals were obtained from methanol solution as very thin, colorless and transparent needles with the long direction along the b-axis. The unit cell parameters determined from rotation and Weissenberg photographs were: a = 16.70, b = 5.93, c = 20.84 Å, β = 91°52′.

The density value of 1.405 g/cc., determined by the flotation method in a benzene-carbon tetrachloride mixture indicated that this crystal contained eight molecules in a unit cell.

The systematic extinctions, hkl when h+k is odd, and h0l when l is odd, showed that the space group was either C2/c or Cc. However, we chose C2/c as the space group of this crystal which has a center of symmetry, and it was confirmed by the Wilson's statistics.

Partial three dimensional intensity data were collected by the multiple-film, equi-

inclination Weissenberg techniques. 1084 independent non-zero intensities were visually estimated and corrected for Lorentz and polarization factors, but no absorption correction was applied. The data were then adjusted to the absolute scale, and normalized structure factor, E , and Σ_2 lists ($|E| \geq 1.75$, probability greater than 97 %) were calculated by the program 'SIGMA' written by Dr. Tamaichi Ashida.

The structure was solved by the use of the symbolic addition method proposed by Karle and Karle. To initiate the symbolic addition procedure, five reflections were selected; two reflections, (8 2 $\overline{17}$) and (3 3 17), for the specification of the origin of the non-primitive space group C2/c, and three reflections, (1 3 16), (8 2 $\overline{14}$) and (7 1 $\overline{18}$), with symbols, a, b and c, respectively[6]. The phases of one hundred and ninety-eight reflections were calculated by hand in terms of the initial signs and symbols. In three dimensional E-map based on the case assuming the signs of a, b and c as all negative, the peaks corresponding to the atomic positions of 5-hydroxytryptophan molecule except hydrogen atoms were obtained. It was shown in the upper half part of Fig.1, in which the contours are drawn at equal intervals with arbitrary scale. Refinement of the structure was carried out by using the block diagonal least squares program 'HBLS IV' on the HITAC 5020E Computer, Tokyo University. After five cycles refinement with anisotropic temperature factors, the R-factor dropped to 0.16. In the composite electron density map of the molecule viewed along b-axis (lower half part in Fig.1), the contours are drawn at interval of 2 e.Å^{-3}, biginning with the 2 e.Å^{-3} contour.

A three dimensional difference Fourier map reveals the appropriate peaks of the hydrogen atom at reasonable positions, and it was confirmed that the molecule of 5-hydroxy-DL-tryptophan exists as the zwitter ionic form in solid state. This molecule is characterized by two planes, one is the indole ring and the others the plane of carboxyl group, which have a dihedral angle of about 40°, while the equivalent angles in L-tryptophan hydrochloride and 3-indolylacetic acid are 70.6° and 62.9°, respectively.

It is quite interesting to compare the conformation about the C(α)-C(β) bond of 5-hydroxy-DL-tryptophan with that of the other related compounds. In this molecule, the orientation of C(β)-C(γ) bond is <u>trans</u> with respect to the C(α)-COO^-. In L-tryptophan hydrochloride, however, this orientation is <u>gauche</u> with respect to both the C(α)-COO^- and C(α)-NH_3^+.

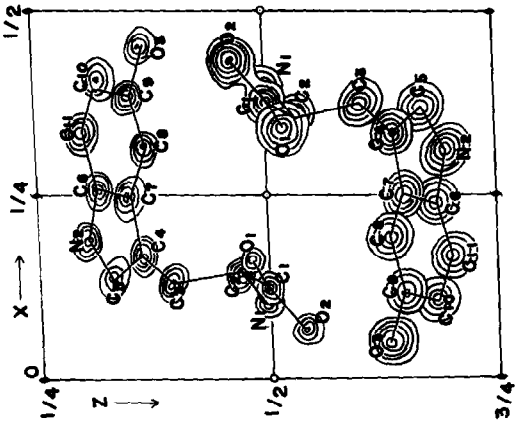

Fig.1.

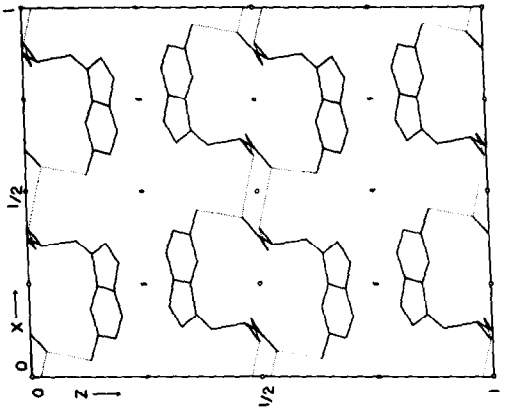

Fig.2.

Fig.2 shows the arrangement of the molecule and the hydrogen bonding system which is indicated by broken lines. Three hydrogen atoms of NH_3^+ group and one hydrogen of hydroxy group may take part in the hydrogen bond formation. However, the distance of N(1)-H---O(3) hydrogen bond is slightly longer than the normal distance, and it is not shown in Fig.2.

The D- and L-forms of 5-hydroxytryptophan are hydrogen-bonded through N-H---O into dimer related by center of symmetry. These dimers are connected through four hydrogen bonds (O-H---O) with neighboured dimers into an infinite chain along the a-axis. The molecules of same form as D to D or L to L are connected with each other through N-H---O bond. The indole rings are mainly stacked with one another by van der Waals force and piled up as a louver along the b-axis. The hydrogen attached to nitrogen in the indole ring is not involved in any hydrogen bonding as in L-tryptophan hydrochloride or 3-indolylacetic acid. On the other hand, it is hydrogen-bonded to oxygen atom of sulphate ion in the case of serotonin.

REFERENCES

1) J.L. Gray, J.T. Tew and H. Jensen, Proc. Soc. exp. Biol. Med., 80, 604 (1952).
Z.M. Bacq, Acta radiol., 41, 48 (1954).
H. Langendorff and R. Koch, Strahlentherapie, 102, 58 (1957).
H. Langendorff and H.J. Melching, Strahlentherapie, 110, 505 (1959).
2) S. Kobayashi, W. Nakamura and H. Eto, Int. J. Rad. Biol., 11, 505 (1966).
3) I.L. Karle, K.S. Dragonette and S.A. Brenner, Acta Cryst., 19, 713 (1965).
4) T. Takigawa, T. Ashida, Y. Sasada and M. Kakudo, Bull. Chem. Soc. Japan, 39, 2369 (1966).
5) I.L. Karle, K. Britts and P. Gum, Acta Cryst., 17, 496 (1964).
6) H. Hauptman and J. Karle, Acta Cryst., 12, 93 (1959).