Studies on the Structure of β-Chitin, I

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Structure of β-Chitin

The structure of β-chitin has been studied in detail. Atomic coordinates in the unit cell are determined. Various positions of O_6 atom have been studied in detail in order to reduce X-ray discrepancies. It is f duonthat O_6 atom lies near 140°,

Introduction

Chitin, a polysaccharide of N-acetyl-D-glucosamine is widely distributed in nature [1]. It occurs in three forms namely α-, β-, γ-chitin [2]. The presence of β-chitin was reported by Lotmar and Picken [3]. The structure of β-chitin proposed by Dweltz [4] was rejected by Carlström [5] on stereochemical grounds. Dweltz [6] proposed the structure of β-chitin of diatom with monoclinic unit cell of dimensions $a = 4.80$ Å, $b = 10.32$ Å (fibre repeat), $c = 9.83$ Å, $\beta = 112°$ and with space group $P2_1$. Blackwell [7] proposed the structure of β-chitin with monoclinic unit cell of dimensions $a = 4.85$ Å, $b = 10.38$ Å (fibre repeat), $c = 9.26$ Å, $\beta = 97.5°$ and with space group $P2_1$.

The present work describes: 1. comparison of two structures of β-chitin proposed by Dweltz [6] and Blackwell [7]. 2. Atomic co ordinates in the unit cell are derived from standard bond length and bond angles of Ramachandran *et al.* [8]. 3. Determination of the coordinates of O_6 atom. 4. Calculations of the structure factors, R-value [9–11] and Φ value [10, 11] after parameter variations.

Determination of Atomic Coordinates in the Unit Cell [12]

The detailed mathematical derivations involved in this procedures are given elsewhere [12]. A brief account for determination of coordinates in unit cell similar to α-chitin is given below.

1. *Determination of the coordinates of glucopyranose ring*

Coordinates representing standard bond lengths and bond angles were determined in the unit cell for the structure proposed by Dweltz [6] and Blackwell [7]. As a first step the average coordinates for general glucopyranose ring proposed by Ramachandran *et al.* [8], was accepted. The following procedure was adopted for the determination of coordinates in the unit cell for both structures [6, 7].

Starting from the atomic coordinates in the reference frame of Ramachandran *et al.* [8] (OXYZ-system I), the coordinates are obtained in a second reference frame with O_4 as origin O′, $O_4 - O_1$ lying along O′Y′ axis and C_5 in the plane of X′Y′Z′ (O′X′Y′Z′-system II, Fig. 1).

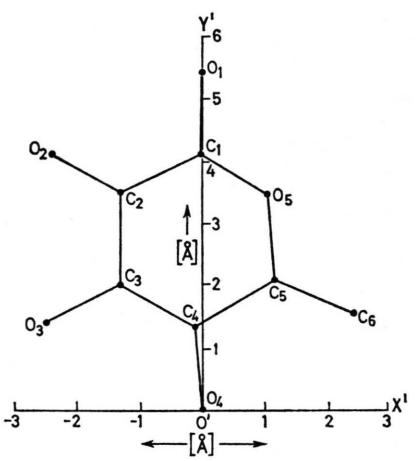

Fig. 1. 001 projection of glucopyranose ring in System II. (O_4 as origin, $O_4 - O_1$ lying along O′Y′ and C_5 in the plane of X′ Y′ Z′.)

Turning to the Blackwell model, we define a coordinates frame III in the identical manner to that used for system II. The transformation of the Ramachandran *et al.* [8] coordinates of system II for

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the atoms of glucopyranose ring define our present structure. Finally inverse transformation was used to change coordinates of system III back to that of the basic system defined by monoclinic unit cell.

2. *Determination of the coordinates of peptide group*

In order to add the standard peptide group of Ooi *et al.* [13], to the glucopyranose ring just obtained, the following procedure was adopted.

We assume the vector $\overline{C_2O_2}$ of the generalized glucopyranose ring to become the direction of $\overline{C_2N_2}$ vector of β-chitin. As a further restriction we assume that the plane of the peptide group contains OX axis of the unit cell (system I). Starting from the atomic coordinates in the reference frame of Ooi *et al.* [13], the coordinates are obtained in the second reference frame with $C_{\alpha i+1}$ as origin, $C_{\alpha i+1}$——N_i lying along the OX and OY lying in the plane of peptide group (system II). Turning to the coordinates of glucopyranose ring, we define a system of coordinates such that the origin O″ lies at C_2, the O″X″ lies along C_2-O_2 and X″O″Y″ plane contains the OX direction of the unit cell (system III). The transformed Ooi *et al.* [13] coordinates when transformed to system III completes our systematic structure of β-chitin. Finally we use inverse transformation to obtain peptide coordinates in the frame of monoclinic unit cell. Similar calculations were carried out for the determination of coordinates of the glucopyranose ring and peptide group for Dweltz [6] model of β-chitin.

3. *Determination of the coordinates of O_6 atom*

The coordinates of O_6 atom were not considered in the Ramachandran *et al.* [8] system as being yet to be determined in the angular position about C_5-C_6 bond. The data from single crystal indicates that the most favourable position is "gt" position [14].

The position of the O_6 atom is *gauche* to C_5-O_5 and *trans* to C_4-O_5 in terms of angular variable describing the position with respect to C_5-C_6 bond. We define Φ' (the angle of rotation) to be zero in 'gt' position, and to increase positively as the plane $C_5-C_6-O_6$ rotates clockwise when viewed along the direction C_5-C_6. The atom C_6 is taken to be the origin of the coordinates and C_5-C_6 parallel to OX, C_4 lying in the plane XOY (Fig. 2).

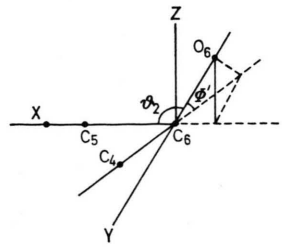


Fig. 2. The position of O_6 atom and angle of rotation (Φ') to be zero in 'gt' position, and to increase positively as the plane $C_5-C_6-O_6$ rotates clockwise when viewed along the direction C_5-C_6. The atom is taken to be origin of coordinates, C_5-C_6 parallel to OX, C_4 lying in plane XOY.

The coordinates for the O_6 atom were calculated for varying values of Φ' for both structures [6, 7]. The coordinates determined for Blackwell [7] and Dweltz [6] model are called R−B system and R−D system respectively. The distance of O_6 atom from the neighbouring atoms for a given value of Φ' in the R−B system was also calculated (Fig. 3). It is interesting to note that the position of O_6 atom in Blackwell and Dweltz model of β-chitin lies in the range of $\Phi' = 290°$ to $310°$.

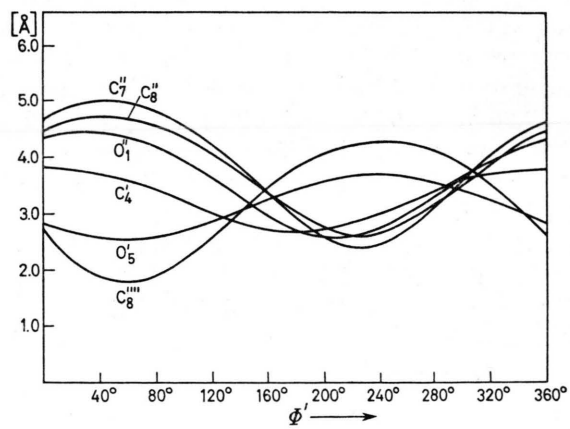


Fig. 3. Distance of the atoms from the O_6 atom for varying values of Φ'.

Results and Discussion

The observed intensities for Dweltz [6] model was reported for only 13 planes and probably owing to this lack of observed intensities, *R*-value was not reported by Dweltz [6]. The calculated *R*-value for Dweltz [6] model was found to be 0.45 and Φ was 141.83. Fig. 4 shows comparision of R−D system and Dweltz [6] model. The calculated and observed structure factors of β-chitin reported by Blackwell [7]

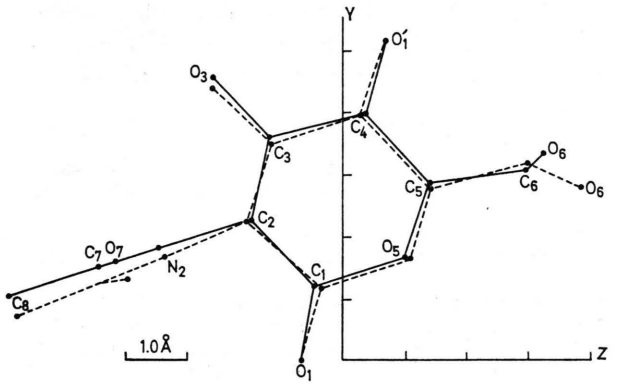


Fig. 4. Projections of N-acetyl-D-glucosamine as determined by Dweltz [6] (shown as dotted line) and as obtained in the present work (shown as continuous lines).

were used to recalculate the R-value and it was found to be 0.36. The calculated value of Φ for Blackwell model was found to be 68.85. Comparision of calculated and observed structure factors for Blackwell model [7] shows large discrepancies for the following planes:

$10\bar{2}$, 102, $11\bar{2}$, 113, $11\bar{4}$, 120, 131 and $13\bar{2}$.

A comparision of Blackwell [7] and Dweltz [6] model indicates that Blackwell's structure is close to the true structure.

Different positions of the O_6 atom which were free from short contacts were obtained for R – B system in the manner described above. The positions of O_6 atom at different Φ' values with respect to neighbouring atoms for R – B system are shown in Fig. 3. Positions of the O_6 atom at Φ' about 140° and between 290° and 320° were found to be free from short contacts. Structure factor calculations were made using coordinates of glucopyranose ring, peptide group and various positions of the O_6 atom. The rotation of O_6 atom has also considerable effect upon R-value and Φ value. A value of $\Phi = 49.68$ was obtained at $\Phi' = 140°$. (Table I shows R-values and Φ values for both Blackwell [7] and Dweltz [6] models.)

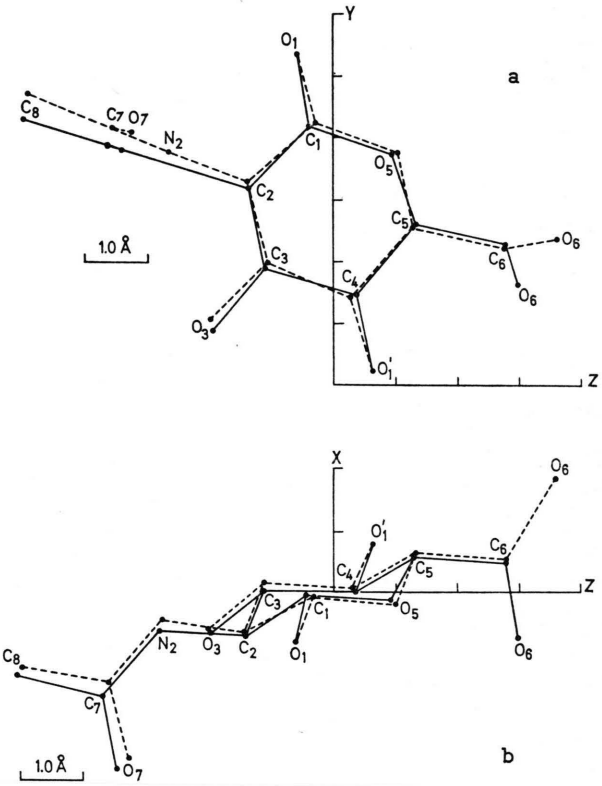
It is clear from Table I that Φ-value for the Dweltz [6] structure is high as compared to Blackwell's structure. Calculation of structure factors for R – B system considering various positions of O_6 atom were carried out and O_6 position at $\Phi' = 140°$ was found to be the best position (R-value = 0.31, $\Phi = 49.68$). It was found that the rotation of O_6 atom about $C_5 - C_6$ bond has considerable effect on the following reflections:

Table I.

	R-Value	Φ-Value
1. Original Blackwell model	0.36	68.85
2. Original Dweltz model	0.45	141.83
3. R – B system ($\Phi'=290°$)	0.36	69.01
4. R – B system ($\Phi'=320°$)	0.39	79.64
5. R – B system ($\Phi'=140°$)	0.31	49.68
6. R – D system ($\Phi'=120°$)	0.44	99.48
7. R – B system ($\Phi'=140°$) 0.2 Å shift in chain direction	0.31	49.81
8. R – B system ($\Phi'=140°$ −0.2 Å shift)	0.31	49.65

(002, $10\bar{1}$), 102, $10\bar{2}$, 112, 113, 124 and 131.

The rotation of O_6 atom about $C_5 - C_6$ bond has also considerable effect on R-value and Φ-value. A shift of 0.2 Å or −0.2 Å along the fibre axis produced little effect on Φ-value and R-value (Figs 5 a and 5 b show comparison between R – B and Blackwell [7] system). In the original model of β-chitin



Figs 5 a and 5 b. Projections of N-acetyl-D-glucosamine unit as determined by Blackwell [7] (shown as dotted line) and as obtained in the present work (shown as continuous lines).

proposed by Blackwell [7] and Dweltz [6], the position of O_6 atom was taken at $\Phi' = 290^\circ$ and 310° respectively. In the Carlström's model of α-chitin, the similar position of the O_6 atom was taken, however according to this work the most favourable position of the O_6 atom is found in the range of $120 - 140^\circ$. It is possible that there are two positions of O_6 atom.

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