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# The Structure of Cellulose by Conformational Analysis. 2. The Cellulose Polymer Chain

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# The Structure of Cellulose by Conformational Analysis. 2. The Cellulose Polymer Chain

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ABSTRACT

Conformational analysis studies on the tertiary structure of cellotetraose, methyl-β-cellotetraoside, and single cellulose chains were carried out by using calculation of van der Waal, H-bond, electrostatic, and torsional energy interactions between the atoms and molecular groups in both the skeleton and side chains of the cellulose polymer. The β-glucosidic linkages connecting two monomers were proved to be in different ($\Phi^\circ$,$\Psi^\circ$) conformations, with different rotational energy barriers and with different H-bond shielding than the β-glucosidic linkages within the cellobiose-like monomers. This confirms the anomalies in the hydrolytic behavior of cellulose reported by other authors. H-bonds and van der Waal's forces were the predominant factors in the fixation of the most favored conformations. The role of H-bonds is again predominant. Single cellulose chains, not in a crystalline network, were found to be in extended helicoidal conformations. These types of conformation are most probable for cellulose in solution and for the cellulose amorphous regions. The conformations most likely to form the crystalline and amorphous regions of native wood cellulose have been indicated pending further study of the crystalline network. Only two models for wood cellulose, namely the "twofold" helix and oscillating "twofold" helix symmetry, appear likely to satisfy the properties of cellulose and the energy balance deduced for single chains.





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## INTRODUCTION

Conformational analysis studies of the structure of cellobiose and methyl-β-cellobioside were carried out in the previous part of this study [1]. A few "primary" conformations of minimum total energy have been identified. As cellobiose and methyl-β-cellobioside are considered to be the structural monomer units of the cellulose polymer, it was thought important to study with the tool of conformational analysis what stable conformations are assumed by these monomers when linked by β-glucosidic bonds to form the cellulose chain.

In the past [2] it has been assumed that the β-glucosidic bonds connecting glucose residues both (i) within a cellobiose-type monomer and (ii) connecting two cellobiose monomers have the same values of ($\Phi^\circ$,$\Psi^\circ$) rotational angles and are in the same ($\Phi^\circ$,$\Psi^\circ$) conformation. This leads to the idea of a cellulose chain in which all the β-glucosidic bonds have the same ($\Phi^\circ$,$\Psi^\circ$) conformation; thus, a cellulose chain composed of monomers of homogeneous configuration. This assumption, which has been readily accepted worldwide, is, however, only an assumption and not a proven fact. It was only based on studies [2] of the structures of cellobiose-type molecules; i.e., on studies of molecules formed only by two connected glucose residues at the time when glucose itself was still considered the monomer unit of cellulose. Thus, it was then not deemed necessary to study the β-glucosidic bonds linking a unit of two glucose residues with the following and preceeding monomer units. This was also considered unnecessary as the conformational study undertaken was based on the determination of the most stable conformation by using only van der Waal's interactions and only taking into account the atoms of the glucose rings.

When van der Waal's interactions and the atoms of glucose rings only are considered, the conformational analysis results have been interpreted as indicating that cellulose is a polymer chain of homogeneous conformation. However, the dual minima found in the work of Rees and Skerrett also suggest the possibility of a polymer chain in inhomogeneous conformation. It has also been shown, in Part 1 of this study [1], that H-bonds between side-chains or groups are predominant in "fixing" cellobiose-like structures in the most favored conformations of minimum total energy. The H-bonds strength and location within the cellobiose-like monomer are very likely to differ from those of the H-bonds around the glucosidic bonds linking two cellobiose-like monomers following each other. Thus, it is also very likely that the ($\Phi^\circ$,$\Psi^\circ$) values of the conformation of minimum total energy of a cellobiose molecule will be greatly different from the ($\Phi^\circ$,$\Psi^\circ$) values of the conformation of minimum total energy of the β-glucosidic bond connecting two cellobiose-like structures following each other along the cellulose chain.

The study presented in this article is concerned with the identification of the most stable minima of total energy, and thus with the most energy stable ($\Phi^\circ$,$\Psi^\circ$) conformation, of the β-glucosidic linkages connecting two cellobiose-like monomer units. Cellobiose-cellobiose, cellobioside-

cellobioside, and also mixed sequences such as cellobioside-cellobiose and cellobiose-cellobioside linking modes of identical and different monomer conformations were studied. The results obtained can give a good idea of the tertiary structure of the cellulose polymer chain.

## EXPERIMENTAL

The computer program as well as the formulas used to compute the various contributions to the conformational energy were the same presented in Part 1 of this study [1]. Again, all the atoms of the cellotetraose-type molecules examined were taken into consideration. The coordinates for the atoms of cellotetraose-type structures had to be generated mathematically. The coordinates for the first and second cellobiose-type monomers (in the conformations of minimum energy) along the length of the cellulose chain had to be generated from the coordinates of cellobiose-type structures at the conformation of minimum total energy obtained during the first part of this study.

The $\beta$-glucosidic bonds, interatomic and bond distances, and angles were maintained consistent to what was obtained for cellobiose-type structure, by rotation, from x-ray diffraction studies. A mathematical system to obtain the coordinates of the second cellobiose-type monomer from the first, which were representative of the structure of cellotetraose-/-oside-like molecules, had to be developed [3]. The mathematical system used for the purpose was as follows:

### Transformation by Rotation and Translation

We consider the following problem: Given the points $P = (x_1,y_1,z_1,)$, $Q = (x_2,y_2,z_2)$, $P' = (x_1',y_1',z_1')$, $Q' = (x_2',y_2',z_2')$ with respect to the XYZ orthogonal coordinate system such that the Euclidean distance between P and Q is equal to that between P' and Q'. Find a transformation which transforms P to P' and Q to Q'.

The sought after rotation is:

$$T_{rot} = \frac{1}{rr'RR'} \begin{bmatrix} z_2' - z_1' & 0 & x_2' - x_1' \\ 0 & r' & 0 \\ x_1' - x_2' & 0 & z_2' - z_1' \end{bmatrix} \longrightarrow$$

$$\begin{bmatrix} R^2 & 0 & 0 \\ 0 & r'r + (y_2' - y_1')(y_2 - y_1) & r(y_2' - y_1') - r'(y_2 - y_1) \\ 0 & r'(y_2 - y_1) - r(y_2' - y_1') & r'r + (y_2' - y_1)(y_2 - y_1) \end{bmatrix} \longrightarrow$$

$$\begin{bmatrix} z_2 - z_1 & 0 & x_1 - x_2 \\ 0 & r & 0 \\ x_2 - x_1 & 0 & z_2 - z_1 \end{bmatrix}$$

with

$$r = [(x_2 - x_1)^2 + (z_2 - z_1)^2]^{\frac{1}{2}}$$

and

$$r' = [(x_2' - x_1')^2 + (z_2' - z_1')^2]^{\frac{1}{2}}$$

$$R \text{ and } R' = [(x_2 - x_1)^2 + (y_2 - y_1)^2 + (z_2 - z_1)^2]^{\frac{1}{2}} =$$

$$[(x_2' - x_1')^2 + (y_2' - y_1')^2 + (z_2' - z_1')^2]^{\frac{1}{2}}$$

respectively.

## The Complete Transformation

Let (x,y,z) be any given point with respect to the XYZ system. Suppose that it becomes (x',y',z') under the complete transformation, then (x',y',z') is obtained as follows:

$$\begin{bmatrix} x' \\ y' \\ z' \end{bmatrix} = T_2^{-1} T_{rot} T_1 \begin{bmatrix} x \\ y \\ z \end{bmatrix} = T_2^{-1} T_{rot} \begin{bmatrix} x - x_1 \\ y - y_1 \\ z - z_1 \end{bmatrix} = T_{rot} \begin{bmatrix} x - x_1 \\ y - y_1 \\ z - z_1 \end{bmatrix} + \begin{bmatrix} x' \\ y' \\ z' \end{bmatrix}$$

or equivalently

$$\begin{bmatrix} x' - x_1' \\ y' - y_1' \\ z' - z_1' \end{bmatrix} = T_{rot} \begin{bmatrix} x - x_1 \\ y - y_1 \\ z - z_1 \end{bmatrix}$$

This rotation formula is the one we used. To conclude, the formulas used for coordinates transformation

$$(x_1, y_1, z_1) \longrightarrow (x_1', y_1', z_1')$$

$$(x_2, y_2, z_2) \longrightarrow (x_2', y_2', z_2')$$

are shown in Table 1. The proof and procedure used to obtain such formulas have already been reported [3].

The angles rotated which influence the minimum of total energy are

TABLE 1. Formulas to Generate New Coordinates

Suppose $(x_1,y_1,z_1) \longrightarrow (x_1',y_1',z_1')$

$(x_2,y_2,z_2) \longrightarrow (x_2',y_1',z_1')$

with distance $x_1x_2$ = distance $x_1'x_2'$

Given (x,y,z) we require coordinates of (x',y',z') such that x to $x_1,x_2$ is the same as x' to $x_1',x_2'$

Then

$$\begin{bmatrix} x' - x_1' \\ y' - y_1' \\ z' - z_1' \end{bmatrix} = \frac{1}{rr'RR'} \cdot A \cdot B \cdot C \cdot \begin{bmatrix} x - x_1 \\ y - y_1 \\ z - z_1 \end{bmatrix}$$

Where

$$A = \begin{bmatrix} z_2' - z_1' & 0 & x_2' - x_1' \\ 0 & r' & 0 \\ x_1' - x_2' & 0 & z_2' - z_1' \end{bmatrix}$$

$$B = \begin{bmatrix} RR' & 0 & 0 \\ 0 & rr' + (y_2 - y_1)(y_2' - y_1') & r(y_2' - y_1') - r'(y_2 - y_1) \\ 0 & r'(y_2 - y_1) - r(y_2' - y_1') & rr' + (y_2 - y_1)(y_2' - y_1') \end{bmatrix}$$

$$C = \begin{bmatrix} z_2 - z_1 & 0 & x_1 - x_2 \\ 0 & r & 0 \\ x_2 - x_1 & 0 & z_2 - z_1 \end{bmatrix}$$

$$r = [(x_2 - x_1)^2 + (z_2 - z_1)^2]^{1/2} \qquad r' = [(x_2' - x_1')^2 + (z_2' - z_1')^2]^{1/2}$$

$$R = [(x_2 - x_1)^2 + (y_2 - y_1)^2 + (z_2 - z_1)^2]^{1/2}$$

$$R' = [(x_2' - x_1')^2 + (y_2' - y_1')^2 + (z_2' - z_1')^2]^{1/2} \quad (R = R')$$

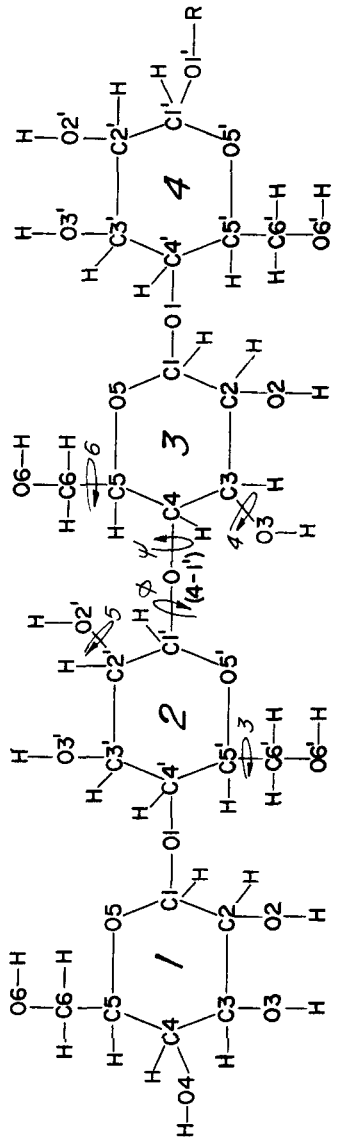

FIG. 1. Positions of bonds rotated in cellotetraose and methyl-$\beta$-cellotetraoside. R = –H when 3-4 glucose rings = cellobiose. R = $-CH_3$ when 3-4 glucose rings = methyl-$\beta$-cellobioside.

the ones shown in Fig. 1, namely the $\Phi^\circ$ and $\Psi^\circ$ of the $\beta$-glucosidic linkage and angle 3', 4', 5', and 6'. (Not to be confused with angles 3, 4, 5, and 6 shown in Part 1 of this study.) The groups which were rotated by these angles are the ones which contribute to the fixation of the $\beta$-glucosidic linkage connecting the two cellobiose-like monomers in its position of minimum total energy. Only the groups which are not fixed by significant H-bonding within each monomer could be rotated to determine the conformation of minimum energy around the linkage of the two monomer units. The conventions used to determine the 0° position were equal to those used in the first part of this study, namely the Ramachandran [1] convention for $\Phi^\circ$,$\Psi^\circ$ and the x-ray crystallographic original position relative to the glucose ring for 3', 4', 5', and 6', for ease of computation.

Both 20° increments and 1° increments maps were computed; the 20° increments maps over a 360° range. Thus, for all the combinations, 104,976 conformations for each monomer-monomer combination were computed. Only "primary" conformations of minimum total energy were obtained for the $\beta$-glucosidic linkage connecting the two cellobiose-type monomer units. The energy values and the $\Phi^\circ$, $\Psi^\circ$, 3', 4', 5', 6' rotational angles defining the conformations of minimum energy obtained are shown in Table 2 for homogeneous conformation monomers.

The H-bond contributions and locations are shown in Table 3. The 20° increments and 1° increments energy maps are shown in Figs. 2 and 3 and the planar projections of the molecules are shown in Figs. 4 and 5. Combinations of different ($\Phi^\circ$,$\Psi^\circ$) conformations of the same monomer, which may be important to the structure of native wood cellulose, are shown in Tables 4 and 5 and Figs. 6, 7, 8, and 9. Combinations of different monomers as, for example, cellobiose followed by methyl-$\beta$-cellobioside, although likely to be of low significance, were also computed and are shown in Tables 6 and 7.

In the determination of the conformations of minimum total energy a first experiment was carried out by computing the ($\Phi^\circ$,$\Psi^\circ$) conformation without changing the position of the side-chains relative to their glucose ring found by x-ray diffraction. The ($\Phi^\circ$,$\Psi^\circ$) conformations found in this case are shown in Table 8. After this, all the following experiments were carried out by rotating all the possible groups which do not help in fixing the intramolecular conformation of the cellobiose and methyl-$\beta$-cellobioside monomers forming the cellotetraose-oside structures.

## RESULTS

Table 2a shows the ($\Phi^\circ$,$\Psi^\circ$) conformations and corresponding total energy minima for the $\beta$-glucosidic linkages between two identical monomers (same type, same internal conformation). These were obtained by fixing the conformation of the two monomers forming the

TABLE 2a. ($\Phi^\circ$,$\Psi^\circ$) Conformations and Values of Total Energy Minima of the Between-Monomers $\beta$-Glucosidic Linkage Connecting Two Consecutive Cellobiose-like Residues Having Identical ($\Phi^\circ$,$\Psi^\circ$) Conformation of Minimum Total Energy and Forming Cellotetraose and Methyl-$\beta$-cellotetraoside (with groups forming intracellobiose-oside H-bonds not allowed to participate to the H-bond)[a]

| Monomer structure and its ($\Phi^\circ$,$\Psi^\circ$) conformation | Conformations of minimum total energy[b] | | | | | |
|---|---|---|---|---|---|---|
| | ($\Phi^\circ$,$\Psi^\circ$) | 3' | 4' | 5' | 6' | Value of total energy minimum (kcal/mol) |
| Cellobiose: | | | | | | |
| a) (32°,138°) | 59°,173° | -29° | 109° | - | - | -3.640 |
| b) (56°,178°) | 59°,173° | -29° | 109° | - | - | -3.641 |
| c) (180°,179°) | 44°,165° | - | - | -87° | - | +1.56 |
| Methyl-$\beta$-cellobioside: | | | | | | |
| d) (-49°,-130°) | -54°,-175° | - | 105° | - | - | +1.264[c] |
| e) (0°,-161°) | -54°,-175° | - | 105° | 168° | - | +1.150[c] |
| f) (168°,177°) | -54°,-175° | - | 104° | 166° | - | +1.082 |

TABLE 2a (continued)

| Monomer structure | Angular shift in vicinal glucose rings at the conformations of minimum: Glucose rings (2-3) (Fig. 1) | Shift from planarity (bent or straight chain); glucose ring (1-2) and (3-4) (Fig. 3) [a] | Shift from planarity (bent or straight chain); glucose ring (2-3) (Fig. 3) [b] | Number of monomers for 360° rotation of helix $\left(\frac{360^\circ}{[a] + [b]}\right)$ |
|---|---|---|---|---|
| Cellobiose: | | | | |
| a) | +232° (-128°) | -10° | +52° | 8.57 |
| b) | +232° (-128°) | +54° | +52° | 3.40 |
| c) | +209° (-151°) | -1° | +29° | 12.86 |
| Methyl-β-cellobioside: | | | | |
| d) | -229° (+131°) | +1° | -49° | 7.5 |
| e) | -229° (+131°) | +19° | -49° | 12 |
| f) | -229° (+131°) | -15° | -49° | 10.59 |

[a] - = 0°, position not varied from relative crystallographic coordinates of side group with glucose ring.
[b] Stabilizing the intercellobioses, -osides conformation.
[c] From what is described in the second footnote, Table 4, Part 1 the actual minima values will be -0.8 and -0.95 kcal/mol for cases d) and e), respectively. See also footnote of Table 2b, Part 2 of this study.

TABLE 2b. Repetition of Table 2a, Where Weak Intracellobiose, -oside H-Bonds Are Allowed to Disappear and Such Groups to Stabilize the Intercellobiose-oside Linkage through New H-Bond Formation

| Monomer structure and it ($\Phi^\circ$,$\Psi^\circ$) conformation | Conformations of minimum total energy | | | | | |
|---|---|---|---|---|---|---|
| | ($\Phi^\circ$,$\Psi^\circ$) | 3' | 4' | 5' | 6' | Value of total energy minimum (kcal/mol) |
| Cellobiose: | | | | | | |
| c) (180°,179°) | 60°,173° | -30° | 108° | - | - | -3.747 |
| Methyl-$\beta$-cellobioside: | | | | | | |
| d) (-49°,-130°) | -58°,-171° | -102° | 98° | - | - | -0.718[a] |
| e) (0°,-161°) | -16°,138° | - | - | 172° | 179° | -1.109[a] |
| f) (168°,177°) | -59°,-171° | -102° | 99° | - | - | -0.812 |

TABLE 2b (continued)

| Monomer structure | Angular shift in vicinal glucose rings at the conformations of minimum: Glucose rings (2-3) (Fig. 1) | Shift from planarity (bent or straight chains); glucose rings (1-2) and (3-4) [a] | Shift from planarity (bent or straight chains); glucose rings (2-3) [b] | Number of monomers for 360° rotation of helix $\left(\frac{360^\circ}{[a] + [b]}\right)$ |
|---|---|---|---|---|
| Cellobiose: | | | | |
| c) | +233° (-127°) | -1° | +53° | 6.92 |
| Methyl-β-cellobioside: | | | | |
| d) | -229° (+131°) | +1° | -49° | 7.5 |
| e) | -154° (-206°) | +19° | +26° | 8.0 |
| f) | -230° (+130°) | -15° | -50° | 5.54 |

[a] From what is described in the second footnote, Table 4, Part 1, the actual minima will be -2.8 and -3.2 kcal/mol for cases d) and e), respectively. This does not influence the relative position of the isoenergetic lines in the energy maps, but must be taken into account in the energy calculations for the packing in a crystalline network of bent-chain conformations (see Part 3 of this study). The formation of a O6---H(02')02' H-bond of value -4.52 is as favored as the set-up presented (energetic value -4.63 kcal/mol).

TABLE 3a. H-Bonds Distribution and Values in the Most Favored Conformations of the β-Glucosidic Linkages Connecting Two Cellobiose-type Monomers to Each Other (glucose rings 2 and 3, Fig. 3) (with groups involved in intracellobiose-oside H-bonding not allowed to participate, Table 2a)

| Cellobiose-cellobiose | | | Methyl-β-cellobioside-methyl-β-cellobioside | | |
|---|---|---|---|---|---|
| Atoms groups | H-bond (kcal-mol) | Contribution (%) | Atoms groups | H-bond (kcal/mol) | Contribution (%) |
| Conformations (32°,138°) duplicated | | | Conformation (-49°,-130°) duplicated | | |
| and | | | 03H(03)– – –05' | -2.625 | 100 |
| (56°,178°) duplicated | | | Conformation | | |
| 03H(03)– – –05' | -4.198 | 52 | (0°,-161°) duplicated | | |
| 06'H(06')– – –03C3 | -3.933 | 48 | 03H(03)– – –05' | -2.633 | 100 |
| Total contribution | -8.131 | 100 | Conformation | | |
| Conformation | | | (168°,177°) duplicated | | |
| (180°,179°) duplicated | | | 03H(03)– – –05' | -2.617 | 100 |
| 06'H(06')– – –03C3 | -2.898 | 85 | | | |
| 03H(03)– – –06'C6' | -0.516 | 15 | | | |
| Total contribution | -3.414 | 100 | | | |

TABLE 3b. H-Bonds Distribution and Values in the Most Favored Conformations of the β-Glucosidic Linkages Connecting Two Cellobiose-type Monomers to Each Other (glucose rings 2 and 3, Fig. 1) (with groups involved in intracellobiose-oside H-bonding allowed to participate where the gain of energy to the inter-monomer linkage is greater than the energy loss of the intramonomer linkage)

| Cellobiose-cellobiose | | | Methyl-β-cellobioside-methyl-β-cellobioside | | |
|---|---|---|---|---|---|
| Atoms groups | H-bond (kcal/mol) | Contribution (%) | Atoms groups | H-bond (kcal/mol) | Contribution (%) |
| Conformation (180°,179°) duplicated | | | Conformation (-49°,-130°) duplicated | | |
| 03H(03)– – –05' | -4.203 | 51 | 03H(03)– – –05' | -2.606 | 55 |
| 06'H(06')– – –03C3 | -3.963 | 49 | 06'H(06')– – –03C3 | -2.121[a] | 45 |
| Total contribution | -8.166 | 100 | Total contribution | -4.727 | 100 |
| | | | Conformation (0°,-161°) duplicated | | |
| | | | 06H(06)– – –02'C2' | -0.093 | 2 |
| | | | 02'H02'– – –06C6 | -5.442[a] | 98 |
| | | | Total contribution | -5.535 | 100 |
| | | | Conformation (168°,177°) duplicated | | |
| | | | 03H(03)– – –05' | -2.610 | 55 |
| | | | 06'H(06')– – –03C3 | -2.105[a] | 45 |
| | | | Total contribution | -4.715 | 100 |

[a]Gains of energy in (-49°,-130°): from -0.27 to -2.121 kcal/mol.
in (0°,-161°): from -0.497 to -5.442 kcal/mol.
in (168°,177°): from - 0.334 to -2.105 kcal/mol.

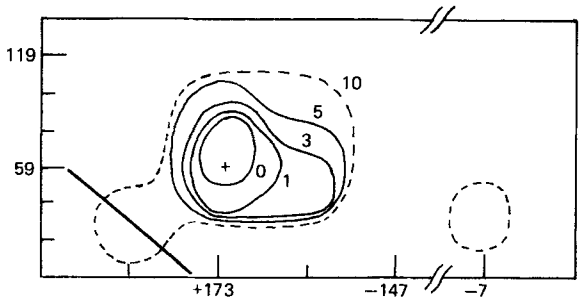


FIG. 2. ($\Phi^\circ$,$\Psi^\circ$) total energy map outline for cellotetraoses composed of (56°,178°) duplicated or (32°,138°) duplicated conformations (Table 2a), or of the 180°,179°) duplicated conformation (Table 2a).



cellotetraose-oside in the internal ($\Phi^\circ$,$\Psi^\circ$) conformations of minimum total energy determined for them in the first part of this study. No chemical group which is involved in H-bonds stabilizing the ($\Phi^\circ$,$\Psi^\circ$) conformation within each monomer was allowed to stabilize the ($\Phi^\circ$,$\Psi^\circ$) conformation of the glucosidic linkage connecting the two monomers. As can be seen in these cases (Table 2a), the ($\Phi^\circ$,$\Psi^\circ$) conformation of the linkage connecting the monomer is very different from the conformation of the similar linkage connecting the two glucose residues within each monomer. By far the most predominant H-bond stabilizing the intermonomers linkage, is the 03H(03)- - -05' formed by the annular oxygen.

However, rotation of some of the groups forming weak H-bonds within each monomer (see Part 1) can form strong H-bonds to further stabilize the conformation of the linkage connecting the two monomers. The ($\Phi^\circ$,$\Psi^\circ$) conformation usually does not change much by rotation of these groups (see Tables 2a and 2b), indicating the inherent stability of the conformations of minimum energy of the intermonomer link which have been determined. The value of the total energy minima are consistently lower. This was done to show that rotations of the groups from their crystallographically determined positions relative to their glucose ring are possible and sometimes energetically advantageous. This will be of importance later when determining which is the more energetically favored cellulose model.

In Tables 3a and 3b all the contributing intramolecular H-bonds in a single chain are listed and their energy contributions given.

As it is likely that in cellulose the cellobiose (or cellobioside) monomers in each single chain are not all in the same internal configuration, combinations of the various monomer conformations were also investigated. In Tables 4 and 5 the relevant data for cellobiose-cellobiose and cellobioside-cellobioside combinations are reported. In Tables 6 and 7 the relevant data of most of the cellobiose-cellobi-

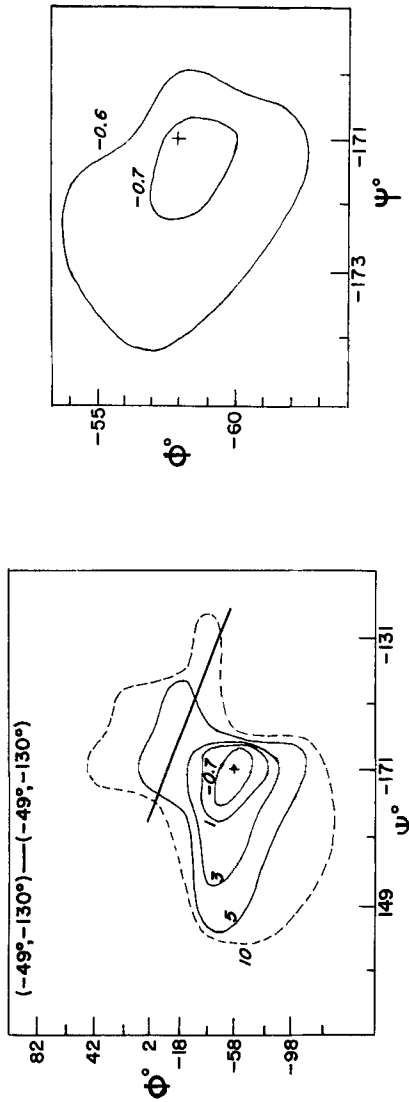


FIG. 3. 20° and 1° increments total energy maps for β-glucosidic linkage connecting two (-49°,-130°) or two (0°,-161°) methyl-β-cellobioside conformations. Energy values in kilocalories.

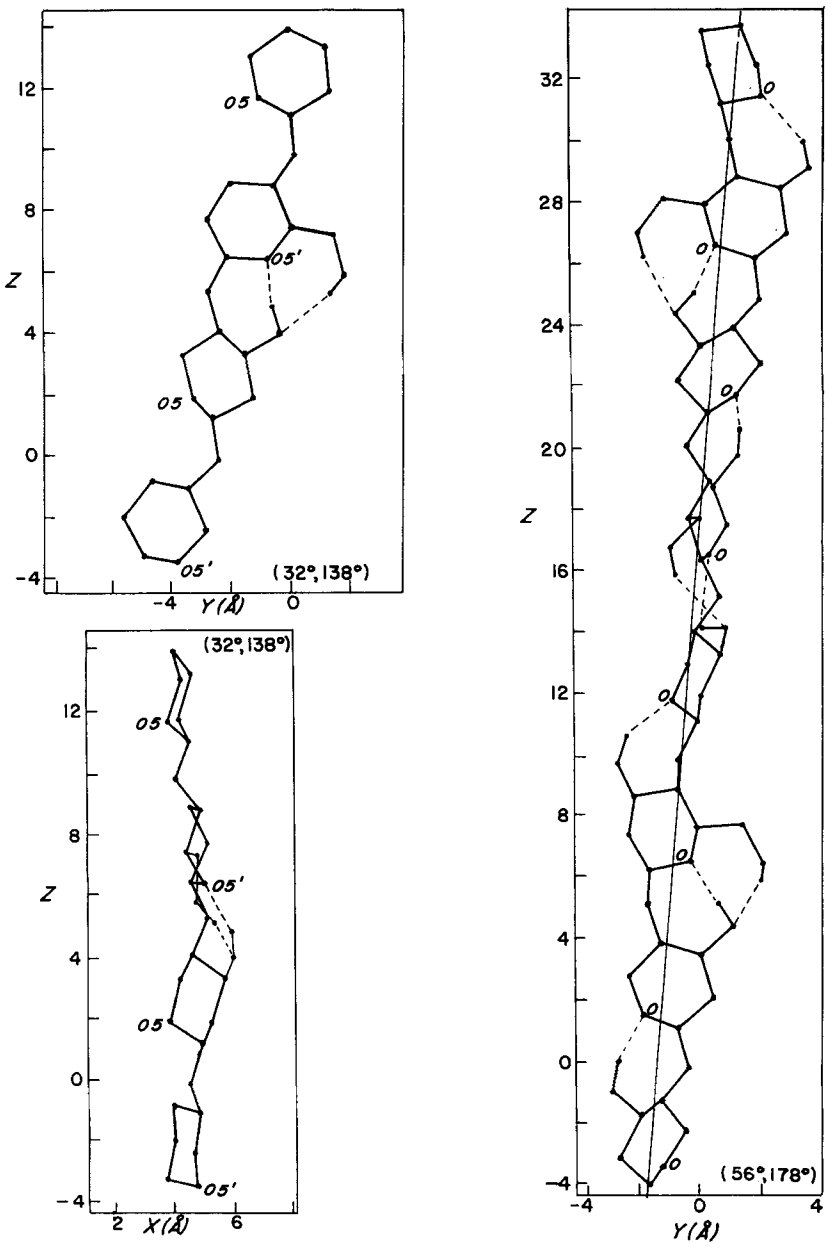


FIG. 4. Plane projections of duplicated (32°,138°) cellobiose conformations. Only the H-bonds formed around the central interconnecting β-glucosidic linkage are reported. The other H-bonds reported are in Part 1 of this study. Plane projection of a sequence of four (56°,178°) cellobiose conformations, showing extended helical conformations. All major H-bonds are reported.

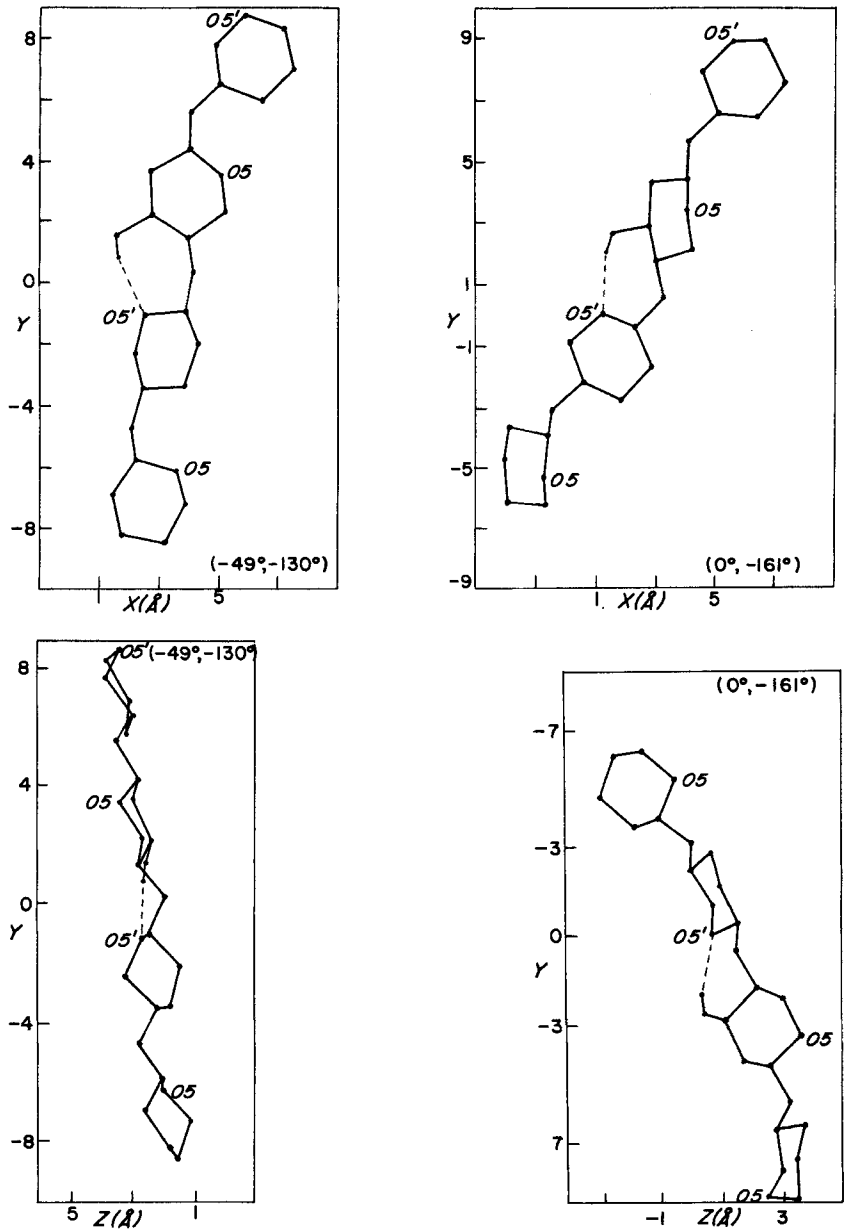


FIG. 5. Plane projections of duplicated (-49°,-130°) and duplicated (0°,-161°) cellobioside couplings showing position of glucose residues and of H-bonds around the interconnecting β-glucosidic linkage.

TABLE 4. ($\Phi^{\circ}$,$\Psi^{\circ}$) Conformations and Values of Total Energy Minima of the Between-Monomers β-Glucosidic Linkage Connecting Two Methyl-β-cellobioside Residues Having Different ($\Phi^{\circ}$,$\Psi^{\circ}$) Conformation of Minimum Total Energy and Forming Methyl-β-cellotetraosides

| Combination no. | 1st cellobioside residue, internal ($\Phi^{\circ}$,$\Psi^{\circ}$) conformation | a | 2nd cellobioside residue, internal ($\Phi^{\circ}$,$\Psi^{\circ}$) conformation | Conformations of minimum total energy of interconnecting β-glucosidic linkages | | | | | Value of total energy minimum (kcal/mol) | Vicinal glucose rings (2-3, Fig. 1) shift in the conformations of minimum | |
|---|---|---|---|---|---|---|---|---|---|---|---|
| | | | | ($\Phi^{\circ}$,$\Psi^{\circ}$) | 3' | 4' | 5' | 6' | | Total angle | Shift from planarity (bent or straight chain) |
| 1 | (-49°,-130°) | A | (0°,-161°) | -54°,-175° | - | 105° | - | - | +1.278 | -229° (+131°) | -49° |
| 2 | (-49°,-130°) | B | (0°,-161°) | -58°,-171° | -102° | 98° | - | - | -0.708 | -229° (+131°) | -49° |
| 3 | (-49°,-130°) | B | (0°,-161°) | -40°,+173° | -104° | - | - | -77° | -1.279 | +133° (-277°) | -47° |
| 4 | (0°,-161°) | A | (-49°,-130°) | -54°,-175° | - | 105° | - | - | +1.263 | -229° (+131°) | -49° |
| 5 | (0°,-161°) | A | (-49°,-130°) | -54°,-175° | - | 105° | 168° | - | +1.136 | -229° (+131°) | -49° |
| 6 | (0°,-161°) | A | (168°,177°) | -40°,-162° | - | - | -130° | -173° | -0.9098 | -202° (+158°) | -22° |
| 7 | (0°,-161°) | A | (168°,177°) | -16°,-139° | - | - | 173° | 178° | -1.1532 | -155° (+205°) | +25° |
| 8 | (0°,-161°) | A | (168°,177°) | -54°,-175° | - | 105° | 168° | - | +1.098 | -229° (+131°) | -49° |
| 9 | (168°,177°) | A | (0°,-161°) | -60°,-173° | - | 106° | -174° | - | +1.199 | -233° (+127°) | -53° |

[a]A = groups involved in intracellobioside H-bonding not allowed to participate to intermonomer H-bonding. B = groups involved in intracellobioside H-bonding were allowed to participate to intermonomer H-bonding as the energy gain in forming intermonomer H-bonds is far greater than the energy loss due to elimination of intramonomer H-bonds.

TABLE 5. H-Bonds Distribution and Values in the Cellotetraoxides by the Mixed Conformations Shown in Table 4

| 1st Methyl-β-cellobioside ⟶ 2nd Methyl-β-cellobioside | | | |
|---|---|---|---|
| Combination no. (Table 4) | H-bonds (rings 2-3; Fig. 1) | | |
| | Atoms groups | kcal/mol | Contribution (%) |
| 1 | 03H(03)– – –05' | -2.625 | 100 |
| 2 | 03H(03)– – –05' | -2.606 | 55 |
| | 06'H(06')– – –03C3 | -2.121 | 45 |
| | Total contribution | -4.727 | 100 |
| 3 | 06H(06)– – –02'C2' | -4.076 | 78 |
| | 06'H(06')– – –03C3 | -1.083 | 21 |
| | 02'H(02')– – –06C6 | -0.046 | 1 |
| | Total contribution | -5.205 | 100 |
| 4 | 03H(03)– – –05' | -2.633 | 100 |
| 5 | 03H(03)– – –05' | -2.633 | 100 |
| 6 | 02'H(02')– – –06C6 | -5.130 | 94 |
| | 03H(03)– – –06'C6' | -0.318 | 6 |
| | Total contribution | -5.448 | 100 |
| 7 | 02'H(02')– – –06C6 | -5.401 | 99 |
| | 06H(06)– – –02'C2' | -0.041 | 1 |
| | Total contribution | -5.443 | 100 |
| 8 | 03H(03)– – –05' | -2.630 | 100 |
| 9 | 03H(03)– – –05' | -2.701 | 100 |

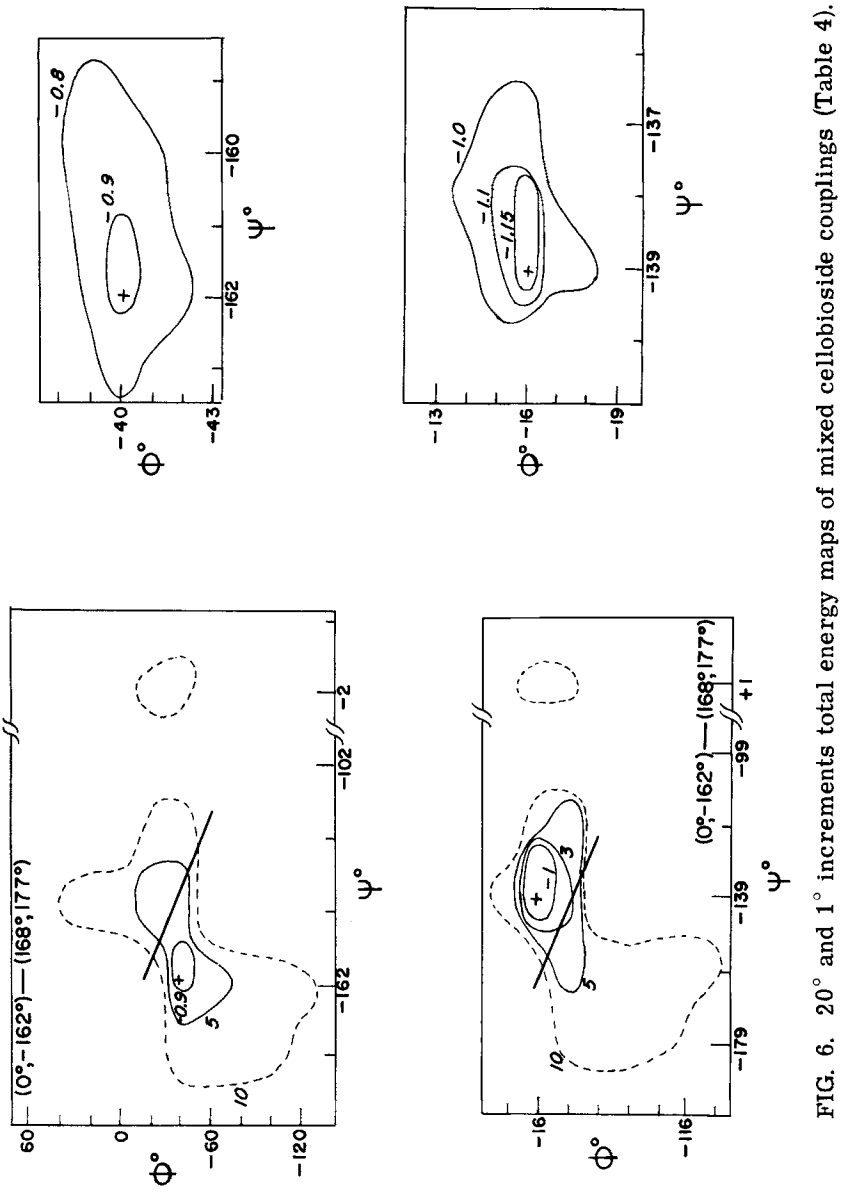


FIG. 6. 20° and 1° increments total energy maps of mixed cellobioside couplings (Table 4).

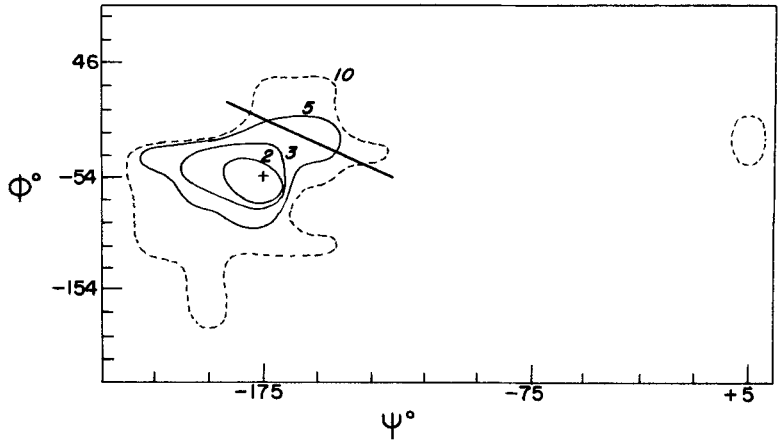


FIG. 7. Total energy maps of all mixed cellobioside confirmations such as (0°,-161°)-(-49°,-130°) having a total angular shift = -229° at the interconnecting β-glucosidic linkage (Table 4, Column 11).



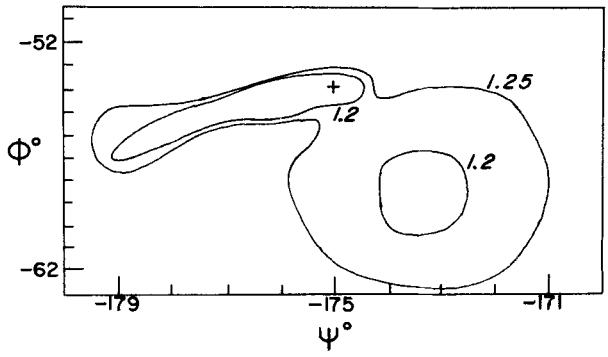


FIG. 8. 1° increments total energy map of (0°,-161°)-(168°,177°) cellotetraoside. Similar minimum arrangements are shown by all the combinations having total angular shift of -229° of the central interconnecting β-glucosidic linkage (valid for Tables 2a and 4; not valid for Table 2b).

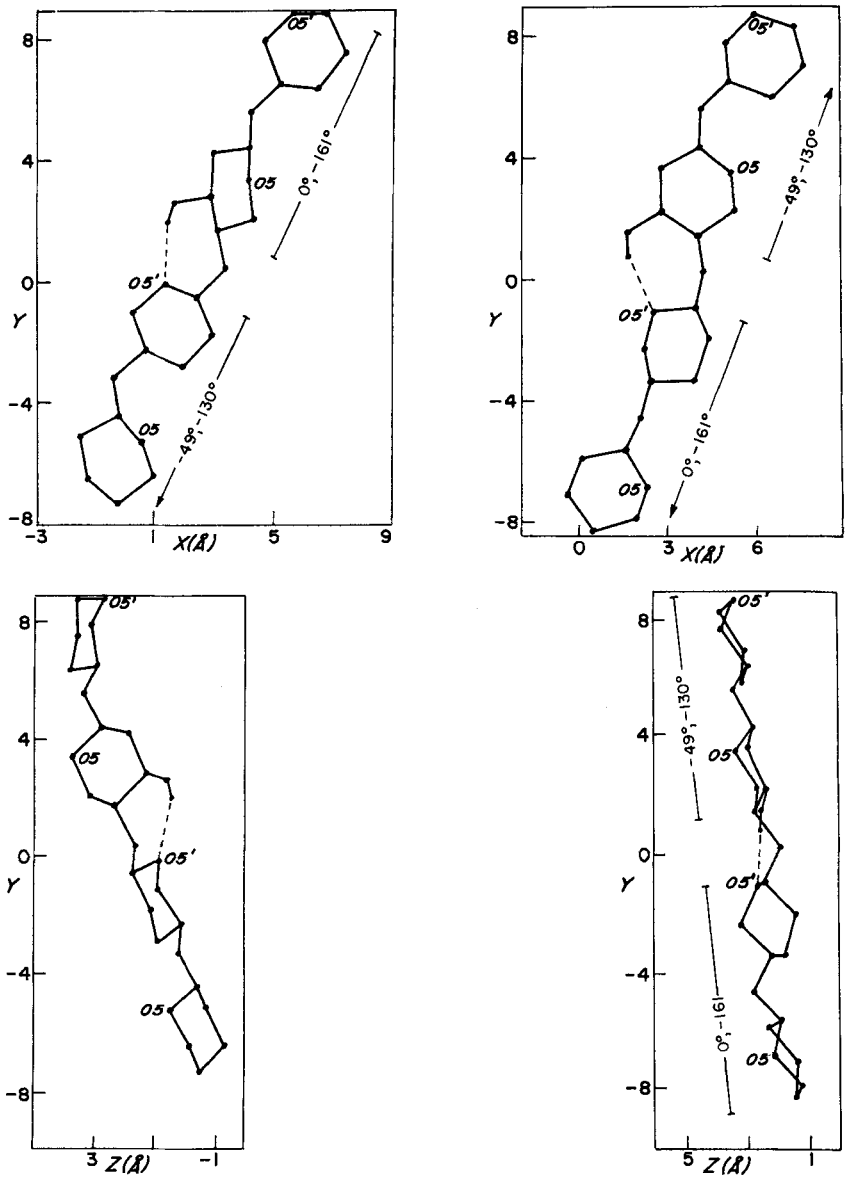


FIG. 9. Plane projections of one mixed cellobioside sequence of particular interest. (-49°,-130°)-(0°,-161°) and (0°,-161°)-(-49°,-130°) describing the possible mixed sequences (-49°,-130°)-(0°,-161°)-(49°,-130°) and (0°,-161°)-(-49°,-130°)-(0°,-161°).

TABLE 6. ($\Phi^\circ$,$\Psi^\circ$) Conformations and Values of Total Energy Minima of Mixed Cellobiose/Methyl-$\beta$-Cellobioside Couplings

| 1st cellobiose conformation (glucose rings 1 and 2, Fig. 3) $\Phi^\circ$,$\Psi^\circ$ | 2nd cellobioside conformation (glucose rings 3 and 4, Fig. 3) $\Phi^\circ$,$\Psi^\circ$ | $\Phi^\circ$,$\Psi^\circ$ | 3' | 4' | 5' | 6' | Total shift angle (rings 2-3, Fig. 3) | Energy minimum (kcal/mol) |
|---|---|---|---|---|---|---|---|---|
| (32°,138°) | (-49°,-130°) | 46°,176° | -91° | 52° | 0° | 0° | +222° (-138°) | 2.816 |
| (32°,138°) | (-49°,-130°) | 169°,-171° | -5° | 123° | 0° | 0° | -2° (+358°) | 2.014 |
| (32°,138°) | (0°,-161°) | 46°,176° | -91° | 52° | 0° | 0° | +222° (-138°) | 2.856 |
| (32°,138°) | (0°,-161°) | 169°,-171° | -5° | 123° | 0° | 0° | -2° (+358°) | 2.045 |
| (56°,178°) | (-49°,-130°) | 45°,176° | -90° | 52° | 0° | 0° | +221° (-139°) | 2.823 |
| (56°,178°) | (-49°,-130°) | 169°,-171° | -5° | 123° | 0° | 0° | -2° (+358°) | 2.000 |
| (56°,178°) | (0°,-161°) | 45°,176° | -90° | 52° | 0° | 0° | +221° (-139°) | 2.864 |
| (56°,178°) | (0°,-161°) | 168°,-171° | -4° | 122° | 0° | 0° | -3° (+357°) | 2.030 |
| (32°,138°) | (0°,-161°) | 62°,175° | 0° | 0° | -71° | 120° | +237° (-123°) | -2.253 |
| (32°,138°) | (0°,-161°) | 172°,-171° | 0° | 0° | -113° | -86° | +1° (-359°) | 0.874 |
| (56°,178°) | (0°,-161°) | 62°,175° | 0° | 0° | -71° | 120° | +237° (-123°) | -2.273 |
| (56°,178°) | (0°,-161°) | 172°,-171° | 0° | 0° | -113° | -86° | +1° (-359°) | - |

TABLE 7. ($\Phi^\circ$,$\Psi^\circ$) Conformations and Values of Total Energy Minima of Mixed Methyl-$\beta$-cellobioside/Cellobiose Couplings

| 1st cellobioside conformation (glucose rings 1 and 2, Fig. 3) $\Phi^\circ$,$\Psi^\circ$ | 2nd cellobiose conformation (glucose rings 3 and 4, Fig. 3) $\Phi^\circ$,$\Psi^\circ$ | $\Phi^\circ$,$\Psi^\circ$ | 3' | 4' | 5' | 6' | Total shift angle (rings 2-3, Fig. 3) | Energy minimum (kcal/mol) |
|---|---|---|---|---|---|---|---|---|
| (-49°,-130°) | (32°,138°) | -17°,150° | -85° | 175° | 0° | 0° | +133° | 0.688 |
| (-49°,-130°) | (32°,138°) | -39°,16° | -112° | 132° | 0° | 0° | -23° | -1.343 |
| (0°,-161°) | (32°,138°) | -17°,149° | 0° | 176° | 0° | 0° | +132° | 0.802 |
| (0°,-161°) | (32°,138°) | -35°,15° | 0° | 130° | 0° | 0° | -20° | 0.785 |
| (-49°,-130°) | (56°,178°) | -18°,150° | -84° | 175° | 0° | 0° | +132° | 0.675 |
| (-49°,-130°) | (56°,178°) | -39°,16° | -113° | 132° | 0° | 0° | -23° | -1.312 |
| (0°,-161°) | (56°,178°) | -20°,160° | 0° | 0° | -106° | 116° | +140° | -0.944 |
| (0°,-161°) | (56°,178°) | -20°,160° | 0° | -127° | -106° | 116° | +140° | -1.210 |
| (0°,-161°) | (56°,178°) | -54°,168° | 0° | 0° | 0° | 116° | +114° | 0.837 |

TABLE 8. ($\Phi^\circ$,$\Psi^\circ$) Conformations and Values of Total Energy Minima of the Between-Monomers $\beta$-Glucosidic Linkage Connecting Two Consecutive Cellobiose-like Residues Having Identical ($\Phi^\circ$,$\Psi^\circ$) Conformation of Minimum Total Energy and Without Allowing Any Rotation of Any Side-Chains from the Positions of Minimum Obtained for the Monomers

| Results from Table 2 (with side-chain rotation) | | With no side-chain rotation | |
|---|---|---|---|
| Conformation | Intermonomers ($\Phi^\circ$,$\Psi^\circ$) | Intermonomers ($\Phi^\circ$,$\Psi^\circ$) | Angular shift; glucose rings (2-3) (Fig. 3) |
| Cellobiose: | | | |
| (32°,138°) | 59°,173° | 46°,164° | +210° (-150°) |
| (56°,178°) | 59°,173° | 46°,165° | +211° (-149°) |
| (180°,179°) | 44°,165° | 52°,170° | +222° (-138°) |
| Methyl-$\beta$-cellobioside: | | | |
| (49°,-130°) | -54°,-175° | -46°,168° | +122° (-238°) |
| (0°,-161°) | -54°,-175° | -46°,168° | +122° (-238°) |
| (168°,177°) | -54°,-175° | -46°,168° | +122° (-238°) |

oside and cellobioside-cellobiose combinations are reported. From Table 4 it is noticeable that the most usual $\Phi^\circ$ and $\Psi^\circ$ angle is again $-229^\circ$ (only cellobioside residues were used for this as the understanding of the structure of cellulose I is our stated aim). This also compares well with the value of 232 to 233° obtained for cellotetraose (Tables 2a and 2b). The difference in total sign -299 and +232° for cellobioside and cellobiose, respectively, indicates only that the two helix are one right-handed and the other left-handed. The 03H(03) ---05 H-bond is again the favorite, although not in all cases (Table 5).

The results in Table 8 (see Experimental) show that the $(\Phi^\circ,\Psi^\circ)$ conformations of minimum energy change considerably if the side-chains are not taken into consideration in the energy calculations. They indicate that the contribution of the side-chains to both the minimization of the potential energy and the optimization of the $(\Phi^\circ,\Psi^\circ)$ values is paramount in any model describing the configuration of the cellulose chain. It also shows that the results obtained by Rees and Skerrett for $(\Phi^\circ,\Psi^\circ)$ with van der Waals functions only for cellobiose are indeed very similar to what we have found with their same approximation (cellobiose, Table 8) but using the total energy instead of van der Waals energy only.

To balance the energy of a chain, the total energy minima of each glucosidic linkage in the chain, both intra- and intermonomers linkages, must be summed. Thus, for the $(-49^\circ,-130^\circ)$ cellobioside conformation in a cellotetraoside residue, the total energy minima will be $-0.607 + 1.264 - 0.607 = +0.050$ kcal/mol (Part 1; this article, Table 2a). The most favorable, thus most probable, energy balance can be taken as $-0.607\ (+0.27) - 0.719 - 0.607\ (+0.27) = -1.392$ kcal/mol (Table 2b) for the most energetically stable conformation. Thus, for a cellobioside-like monomer within the body of an uninterrupted chain sequence, the balance of energy, for a $(-49^\circ,-130^\circ)$ conformation, will be $-0.607\ (+0.27) - 0.718 = -1.055$ kcal/mol.

By straining two cellotetraoside parallel molecules into "twofold" helix symmetry, from a $(-49^\circ,-130^\circ)$ conformation, with the minimum waste of energy, it is possible to calculate that the energy shifts to a value of +4.19 kcal/mol for each cellotetraoside. The calculation is done from the values in the energy maps of both the monomer and intermonomers linkage (Part 1, Figs. 2 and 3, and this article, Figs. 2 and 3) (observe in the energy maps the minimum difference in energy given by the distance between the straight line where "twofold" helix symmetries lie and energy minimum). The value of +4.19 kcal/mol indicates that straining the molecule into a "twofold" helix conformation causes a minimum loss of energy per each chain of $4.19 - (-1.392) = 5.582$ kcal/mol. Thus, crystalline packing will occur only if by combining the two cellotetraoside chains, the gain in total energy obtained is higher than the $5.582 + 5.582 = 11.164$ kcal/mol loss incurred in forcing each molecule into a "twofold" helix symmetry. Due to the low energy gain required, it is fair to assume that a cellulose chain formed of $(-49^\circ,-130^\circ)$ cellobioside-like monomers is indeed

likely to form a crystalline network. Conversely, the (0°,-161°) primary conformation has an energy balance for cellotetraoside of +0.04 - 1.109 + 0.04 = -1.029 kcal/mol. Forcing the cellotetraoside into bent-chain symmetry, the energy loss is approximately 11 to 12 kcal/mol per chain. Thus, a total energy gain well above 20 kcal/mol is needed to pack in a crystalline network two cellotetraosides in (0°,-161°) conformation strained into a bent-chain. The energy gain required appears to be too high to be satisfied by the interchains H-bonds that can be formed. Consequently, a chain originally formed by monomers in (0°,-161°) conformation is quite likely not to be present in crystalline cellulose, but is probably the most likely component of the amorphous regions. The same reasoning also applies to the secondary conformation.

If mixed conformations do exist such as (0°,-161°) followed by (-49°,-130°) or vice versa, the ($\Phi$°,$\Psi$°) and energy values of the (0°,-161°) - (-49°,-130°) and of the (-49°,-130°) - (0°,-161°) sequences may sometimes be different. This is due to the different side-chains available for contribution to the intermonomer glucosidic linkage. Thus, both types of combinations were investigated (see Tables 4, 5, 6, and 7; Figs. 6, 7, 8, and 9).

It must be noticed that in the (-49°,-130°) - (-49°,-130°) configuration a strong 02'H(02')- - -06 H-bond as theorized by other authors can indeed be formed. However, the H-bonds and van der Waals forces pattern reported in the tables give better stabilization in a single chain. We have already found, in the third part of this study, that the situation is reversed in bent-chain packing in a crystalline network, thus confirming the results obtained by x-ray analysis.

## DISCUSSION

The results shown indicate clearly that the glycosidic linkages in cellulose have different conformations. It is certain that even when the glycosidic linkages within each monomer have the same conformation, they differ from the glycosidic linkages connecting two monomers with each other. Thus, it appears that the cellulose chain, at least in the amorphous regions, is composed of monomers which have different internal conformations. Thus, cellulose is likely to be a chain of heterogeneous conformation. The difference in the morphology of the glycosidic linkages along the chain is certainly one of the reasons, possibly the main reason, for the differences in susceptibility to hydrolytic attack of different regions of cellulose. In this respect the conformational analysis strongly confirms the indications already obtained by Raman spectroscopy [4-7].

It is also clear that in chains composed of cellobiose or cellobioside monomers, the glycosidic linkage does not exist in a conformation consistent with "twofold" helix symmetry. This statement is certainly valid for isolated cellulose chains which are not combined with other chains to form a crystalline network. In the case of a chain in a

crystalline network, the same might also apply. Cellobioside-based chains correspond to a right-handed departure from the twofold helix line while cellobiose-based chains correspond to a left-handed departure from this line. Considering the differences in sign of $\Phi^\circ$ and $\Psi^\circ$ which represent this fact, the helix structures formed by the two monomers are amazingly similar (see Table 2a and Figs. 4 and 5).

These findings do not preclude the existence of a "twofold" helix conformation in the crystalline portion of cellulose. They only indicate that if the "twofold" helix conformation of cellulose is correct, at least for isolated chains, the glycosidic linkages will not be in the conformation of most favorable potential energy. This means that to the increase in potential energy caused by shifting the glycosidic linkage from one of the conformations of minimum energy to a "twofold" helix symmetry must correspond an at least equivalent or greater decrease in potential energy due to interactions between parallel chains in the packing of the crystalline network (see Results). This is probable as the H-bonds which appear to contribute strongly and predominantly to the minimization of the potential energy of the chain are also the main type of connection between parallel chains. If and when this happens, the pattern of H-bonding surrounding the glycosidic linkages will not change much from that obtained for the various conformations of minimum energy. Certainly the weaker H-bonds might disappear and the stronger ones may weaken, but the higher steric hindrance of a conformation in relation to a less sterically hindered one will not disappear.

Again the heterogenity of the glycosidic linkages, and therefore the difference in steric hindrance between conformations with different H-bonds "casing," will contribute to the differences in the rate of hydrolytic attack of the different zones in the cellulose chain.

Of the six conformations of minimum energy for the monomers (three for cellobiose, three for cellobioside), the four "primary" conformations are by far the most likely to exist. Thus, a cellobioside-based chain not in "twofold" helix symmetry can be formed of (i) monomers all in one primary conformation only and (ii) monomers of both types of primary conformation forming regular or random sequences. The quantity of the two monomers in the sequence may well be very different. Occasional monomers having a "secondary" minimum energy conformation may also be present.

These observations lead to the conclusion that not only one most favored "twofold" helix conformation of cellulose may exist, but instead several types. (i) The "twofold" helix symmetry is not the position of minimum energy of the glucosidic bonds, and stabilization is achieved through chains packing in a network. (ii) The different conformations of minimum, once forced into a "twofold" helix symmetry, will partly conserve their different H-bonds casing around the glycosidic linkage, thus remaining different even if all are in "twofold" helix symmetry, several types of "twofold" helix conformations may exist. The difference is in the H-bond pattern within each monomer and not in the H-bond pattern around the glycoside linkage connecting

two monomers. The latter might show differences but these are less likely and of very much lower frequency. Thus, just taking into account the primary conformations of minimum energy of cellobioside for cellulose I, we may well have "twofold" helix conformations retaining the H-bond casing of (i) the minimum conformation (0°,-161°) only, (ii) the minimum conformation (-49°,-130°) only, and (iii) a variety of bent-chain conformations in which the (0°,-161°) and (-49°, -130°) are in regular or random sequence and in different abundance.

Thus, it may well not be correct to talk about a "twofold" helix conformation. The problem is: Which "twofold" helix conformation? It is possible that only one or two of them or that several "twofold" helix conformations do exist. However, it is likely that one type of packing is energetically more favorable than all the others and thus only one type of "twofold" helix conformation may exist in the crystalline zones of cellulose. It is also likely that only one conformation strain into a "twofold" helix symmetry is so energetically favorable as to allow formation of a crystalline network.

It is interesting to see, from Table 2a, the period of the helix of the chains formed by the various conformations. It must be pointed out that the same chain can be described as a function of two helixes. That is, the helix is formed by the total ($\Phi°$,$\Psi°$) shift and the helix is formed by the shift of the chain from ribbonlike planarity. Thus, the molecules formed by the duplicated cellobioside conformation (0°,-161°) (see Table 2a) has the first helix in which the number of monomers with a 360° rotation is 360°/[-161°+(-229°)] = 0.92 monomers and the second helix, which is given by deviation from planarity, in which the number of monomers needed to have a 360° rotation is 360°/[+19°+ (-49°)] = 12 monomers (24 glucose rings, thus 124 Å).

To give an idea of the shape and tightness of these helixes, the tighter helix, that of the cellobiose (56°,178°) conformation, is shown in Fig. 4. As can be seen, this is an extended helix even if it is the tightest of them all. Thus, the helix given by a conformation such as the cellobioside (0°,-161°) is indeed very extended and not easily noticeable when short-chain residues are depicted (see Fig. 5).

It is possible that, in contrast to the pressure of a "twofold" helix symmetry, monomers of different helicoidal conformations are combined in such a way as to give a chain nearly flat in which the mean planes of the glucose rings in the sequence shift from the ribbonlike planarity of the bent-chain symmetry (of, say, a maximum of 30 to 40°). By combining conformations of total -n° with conformations of total +n° (see tables) per monomer, or simply monomers of total -n° with intermonomer glycosidic linkages of total +n° in prearranged or random sequences, this may well be possible.

A chain of this type could still form some interchain H-bonds in the crystallite packing, producing further stabilization. The interchain H-bonds could, however, be weaker than those formed by a "twofold" helix conformation. The important point is: Would the energy balance of a chain like this, which is composed of a more stable sequence but is less stabilized by the interchain H-bonds than the "twofold" helix

structure, be more or less favorable than "twofold" helix structures composed of a less stable sequence, but stabilized considerably more by interchain H-bonds?

At this stage it is not possible to answer such a question. The effect on the total energy balance of chains packing in a crystalline network will be computed and discussed in the next and last article of this series. We feel, however, that the chances of the existence of this type of structure may be good only for chain zones of short length and the likelihood of this conformation mixture explaining the structure of all crystalline cellulose are fairly remote.

These two models proposed in this article all explain the pronounced swelling and stress anisotropy, low extensibility, high strength, density characteristics, and birifrigence of cellulose. All two models agree with (i) the distribution of fragments of different DP obtained in cutting experiments by Muggli and Mühlethaler [17, 18], (ii) the theory of chain dislocation of Mühlethaler [19], and (iii) the theory of Rowlands and Roberts [20, 21]. The subelementary and elementary fibrils are thus continuous plainly extended or extended helicoidal chains of nearly perfect crystallinity in which the amorphous regions are of similar shape and morphology to the more crystalline ones, but with a lower frequency of interchain H-bonding which renders them more susceptible to hydrolytic attack. Thus, the ideas of Kitaigorodskii and Tsvankin [27, 28] of the amorphous region of cellulose, namely of a phase homogeneous with the crystalline regions, but of poorer tridimensional order in which the cellulose chains are arranged in the regularly recurring nodes of the lattice but display a certain shift (small) with respect to these nodes, also agree with the two models which are possible according to this conformational analysis. They are also consistent with a certain periodicity found along the cellulose network for crystalline and noncrystalline regions are proposed by Hess [29] and with x-ray crystallography data from several authors [13, 16, 30, 31].

The folded chain structure proposed by Manley [8, 9], Bittiger [10], and others [11, 12] has been severely criticized as regards electron microscope analysis of anatomical features [22], mechanical properties [23, 24], degree of polymerization tests [17], and staining techniques [25, 26]. While it may be possible by stabilization due to distortion of the pyranose rings as advocated by Melberg and Rasmussen [32], it is definitely not possible with an undistorted pyranose ring and H-bond stabilization especially due to severe geometrical problems arising at the folding points. However, even by distortion of the pyranose rings, it is energetically less stable than the other advocated conformations and thus its existence is quite unlikely.

## CONCLUSIONS

1. H-bonds are the predominant stabilizing and fixing force of the structure of cellulose.

2. The β-glucosidic linkage within a cellobiose-like monomer always has a different conformation than the β-glucosidic linkages that connect the monomer to the preceeding and following monomers along the cellulose chain.
3. Different monomer and monomer-connecting conformations have different stability and different H-bond "casing." This and the preceeding point are important contributing causes to the differences in the rate of hydrolytic attack observed in different cellulose regions.
4. Isolated cellulose chains, which are not in a crystalline network, do not exist in "twofold" helix symmetry but are in extended-helix symmetry.
5. Only two models of cellulose can be proposed that are stable as regards their potential energy. Folded-chain symmetry is energentically very unstable and is definitely not present in the structure of Cellulose I. The two models possible are:
   (i) A "forced" "twofold" helix symmetry in which the loss of energetic stability due to $(\Phi^\circ,\Psi^\circ)$ rotation from the conformations of minimum energy is compensated for by equivalent or greater gains in energetic stability through strong H-bonding between parallel chains. In this respect not only one but several "twofold" helix conformations may be possible due to the differences in H-bond "casing" around the glycosidic linkages along the chain. Such differences can also account for crystalline and noncrystalline regions.
   (ii) An "imperfect" "twofold" helix symmetry in which the mean planes of the glucose rings following each other in the chain sequence can form moderate angles (oscillate) around the "planarity" of a proper "twofold" helix model. Here, too, different conformations of the glycosidic linkages are present.

We do not favor any of the two models presented over the other, although a "twofold" helix model also appears to us to be the most likely to exist in the crystalline region of cellulose. The subject of the last article of this series, in which conformational analysis of the crystallographic networks proposed will be carried out, will help to decide which of the three models is really the most probable.

## ACKNOWLEDGMENT

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## REFERENCES


[1] A. Pizzi and N. Eaton, J. Macromol. Sci.–Chem., A21, 1443 (1984).
[2] D. A. Rees and R. J. Skerrett, Carbohydr. Res., 7, 334 (1968).
[3] L. Rolfes, Private Communications, 1982; N. Eaton and A. Pizzi, Bonds Conformational Analysis Programme Manual, CSIR Hout Report, Pretoria, South Africa, 1983.
[4] R. H. Atalla, "Conformational Effects in the Hydrolysis of Cellulose, Chapter 3 in Hydrolysis of Cellulose: Mechanisms of Enzymatic and Acid Catalysis, (R. D. Brown and L. Jurasek, eds.), Advances in chemistry Series No. 181, Americal Chemical Society, Washington, D.C., 1979.
[5] R. H. Atalla and B. E. Dimick, Carbohydr. Res., 39, C1 (1975).
[6] R. H. Atalla, Appl. Polym. Symp., 28, 659 (1976).
[7] R. H. Atalla, B. E. Dimick, and S. C. Nagel, in Cellulose Chemistry and Technology, American Chemical Society Symp. Ser., Vol. 48, 1977, p. 30.
[8] R. S. J. Manley, Nature, 204, 1155 (1964).
[9] R. S. J. Manley, J. Polym. Sci., Part A-2, 9, 1025 (1971).
[10] H. Bittiger, E. Husemann, and A. Kuppel, J. Polym. Sci., Part C, 28, 45 (1969).
[11] S. K. Asunmaa, Tappi, 49, 319 (1966).
[12] H. Marx-Figini and G. V. Schulz, Biochim. Biophys. Acta, 112, 81 (1966).
[13] H. Mark, Chem. Rev., 26, 169 (1940).
[14] K. H. Meyer and H. Mark, Ber., 61, 593 (1928).
[15] K. H. Meyer and H. Mark, Cellulosechemie, 9, 61 (1928).
[16] K. H. Meyer and L. Misch, Helv. Chim. Acta, 20, 232 (1937).
[17] R. Muggli, Cellul. Chem. Technol., 2, 549 (1969).
[18] R. Muggli, H. G. Elias, and K. Mühlethaler, Makromol. Chem., 121, 290 (1969).
[19] K. Mühlethaler, J. Polym. Sci., Part C, 28, 305 (1969).
[20] S. P. Rowland and E. J. Roberts, J. Polym. Sci., Polym. Chem. Ed., 10, 2447 (1972).
[21] S. P. Rowland, E. J. Roberts, J. L. Bose, and C. P. Wade, J. Polym. Sci., Part A-1, 9, 1623 (1971).
[22] R. B. Hanna and W. A. Côte Jr., Cytobiologie, 10, 102 (1974).
[23] P. Gillis, R. Mark, and R. Tang, J. Mater. Sci., 4, 1003 (1969).
[24] R. E. Mark, J. Polym. Sci., Part C, 36, 292 (1971).
[25] W. Franke and B. Ermen, Z. Naturforsch., 24b, 918 (1969).
[26] W. Franke and H. Falk, Ibid., 23b, 272 (1968).
[27] N. I. Nikitin, The Chemistry of Cellulose and Wood, Israel Programme for Scientific Translations, Jerusalem, 1966; Translated from original book in Russian, Moscow, 1962.
[28] A. I. Kitaigorodskii and D. Ya. Tsvankin, Vysokomol. Soedin., 1, 269 (1959).

[29] K. Hess, H. Mahl, and E. Gutter, Kolloid Z., 155, 1 (1957).
[30] R. O. Herzog and W. Jancke, Z. Phys., 3, 196 (1920).
[31] F. Shafizadeh and G. D. MaGinnis, Adv. Carbohyd. Chem. Biochem., 26, 297 (1971).
[32] S. Melberg and K. Rasmussen, Carbohydr. Res., 71, 25 (1979).

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