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A vibrating flexible chain in a molecular cage: crystal structure of the complex cyclomaltoheptaose (β-cyclodextrin)–1,4-butanediol·$6.25H_2O$*

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ABSTRACT

A single crystal X-ray diffraction study of the title complex carried out at room temperature revealed space group $P2_1$, a = 21.199(12), b = 9.973(3), c = 15.271(8) Å, β = 110.87(3)°, V = 3017(3) Å^3, 4681 unique reflections with $F_o > 1\ \sigma(F_o)$. The structure was refined to R = 0.069, resolution $\lambda/2\sin\theta_{max}$ = 0.89 Å. The crystal packing is of the cage type and is isomorphous to that of β-cyclodextrin (βCD) dodecahydrate. One 1,4-butanediol and ~ 1.25 water molecules are enclosed in each βCD cavity. The hydroxyl groups of the 1,4-butanediol molecule are located at each end of the cavity and form hydrogen bonds with neighboring water and βCD molecules. The flexible $(CH_2)_4$ moiety vibrates extensively in the central part of the cavity. Water molecules and hydroxyl groups are chelated between O-6 and O-5 of at least five glucose residues.

INTRODUCTION

The cyclodextrins (CDs, cyclomalto-oligosaccharides) are cyclic oligosaccharides consisting of six (αCD), seven (βCD), eight (γCD), and nine (δCD) α-D-glucose residues with (1→4) linkages. The CDs form inclusion complexes with numerous molecules of suitable size and have found industrial applications[1,2]. Many of these complexes can be crystallised from aqueous solution and have been studied by X-ray and neutron diffraction methods. Usually, the guest molecules enclosed in the CD cavities are more or less disordered and, except for αCD, few ordered crystal structures have been reported. Since the CD molecules carry three hydroxyl groups per glucose residue and the crystal structures always contain several hydration water molecules, numerous hydrogen bonds are formed. Thus, CD complexes can serve as models for biological systems in order to study hydrogen-bonding networks, and structural and dynamic properties of the guest molecules in the complexes (ref. 3 and references therein).

We now report the X-ray crystal structure of the hexahydrated inclusion complex of βCD–1,4-butanediol. The crystals are isomorphous with those of βCD dodecahydrate[4] and βCD–ethanol octahydrate[3,5] with a cage-type packing. The good crystal-

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* Topography of Cyclodextrin Inclusion Complexes, Part 29. For Part 28, see ref 18.
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lographic resolution allows the study of the structural, and to some degree the dynamic, properties of a partly polar and partly apolar molecule that is enclosed in a molecular cavity.

EXPERIMENTAL

Single crystals were prepared by slow cooling of concentrated solutions of βCD in 2:3 1,4-butanediol–H_2O from 70°. Two sets of data were collected about 1 and 18 weeks after crystallisation. For the latter, a prism of dimensions $\sim 0.5 \times 0.3 \times 0.2$ mm^3 was extracted from the batch tube and sealed in a glass capillary together with some mother liquor. X-Ray experiments were performed on a Turbo-CAD 4 diffractometer (Enraf Nonius) with Ni-filtered Cu-K_α radiation (λ 1.542 Å) from a generator with a rotating anode. The space group is monoclinic $P2_1$, $Z = 2$ (two formula units in the unit cell), and the cell constants (determined from the positions of 18 reflections) are $a = 21.199(12)$, $b = 9.973(3)$, $c = 15.271(8)$ Å, $\beta = 110.87(3)$°, and $V = 3017(3)$ Å^3, with the linear absorption coefficient $\mu_{calc} = 10.4$ cm^{-1}. The cell setting with $a > c$ was chosen as in the isomorphous structures[4,5]. 12201 reflection intensities were measured at room temperature in the $\omega/2\theta$ scan mode with background determinations on both sides of each scan. Intensities were corrected for background, Lorentz, polarisation, and absorption effects (ψ scan method[6]). Symmetry-equivalent reflections were merged to yield a set of 4710 unique data, of which only 29 had values $F_o < 1\,\sigma(F_o)$ ($R_{merge}(F) = 3.4\%$). The data were complete to $2\theta_{max} = 120°$ (the limit for 2θ of the diffractometer used), corresponding to a nominal resolution of $\lambda/2\sin\,\theta_{max} = 0.89$ Å.

Determination and refinement of the structure. — The similarity of the cell constants suggested that the molecular packing in the βCD–1,4-butanediol complex is isomorphous to that of βCD-hydrate[4]. Atomic co-ordinates of the βCD molecule in the latter structure were used to initiate least-squares refinement (SHELX 76[7], function minimised $\Sigma w(|F_o| - |F_c|)^2$, $w = 1.0$ for all reflections, guided by the computer graphics program[8,9] FRODO, version[10] E 2.3, which was used to display electron density maps prepared with the CCP4[11] program package). Several cycles of consecutive least-squares refinement and difference Fourier maps revealed the positions of one 1,4-butanediol molecule, and of five fully occupied and three partially occupied water sites ($\Sigma_{occ} \sim 6.25$) in the asymmetric unit. For the βCD molecule, the H atoms bound to C were placed in their calculated ideal positions. Several hydroxyl and water H atoms were located in the difference-Fourier maps, but since a hydrogen-bonding system could not be assigned unambigously and reliably, they were not included in the final refinement cycles. Refinement of the 1,4-butanediol molecule was complicated by the fact that, for each O and C atom, there was an individual electron-density peak, but some had irregular shapes suggesting extensive vibrations and/or disorder over closely neighboring sites. Attempts to describe these positions as two or more alternative sites, or to fit partially occupied molecules with ideal covalent geometry and different conformations into the electron density, were not satisfactory which might be associated with the limited resolution of ~ 0.9 Å. Finally, each of the 1,4-butanediol atoms was refined as a single atom, which may represent the mean position of several closely neighboring sites. No geometrical constraints were used. Anisotropic refinement converged with $R = 0.069$ for the 4681 reflections with $F_o > 1\,\sigma\,(F_o)$*.

RESULTS AND DISCUSSION

The superscript numbers in the labelling of atoms designate the glucose residues, *e.g.*, C-1^2 indicates C-1 of glucose residue 2 of the βCD molecule.

Crystal packing. — The βCD molecules are arranged in a herring-bone fashion (Fig. 1) so that the cavity of each macrocycle is closed at each end by neighboring βCD molecules. Similar cage-type packing has been observed for several other complexes of βCD with small guests (water[4], methanol[13], HI[13], ethanol[3,5], benzyl alcohol[14], K^{+}[15]). The cavity of each βCD contains one 1,4-butanediol and 1.25 water molecules, *i.e.*, about one out of four βCD molecules in the crystal includes two water molecules (W-6 and W-7); the other three each contain only one (W-6). Five water molecules (W-1 to W-5) per asymmetric unit are located between the macrocycles; four are ordered and one is disordered over two sites (W3-A occupancy ~ 0.80, W-3B occupancy ~ 0.20).

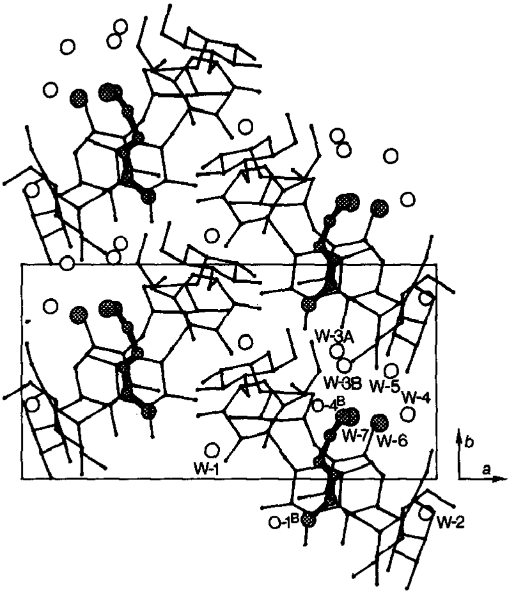

Fig. 1. Crystal packing of the βCD–1,4-butanediol·6.25H_2O complex, viewed along the *c* axis. Shaded molecules are enclosed in the βCD cavities.

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* Lists of fractional atomic co-ordinates, anisotropic and equivalent isotropic thermal parameters for all refined atoms, bond lengths, bond angles, and a selection of torsion angles for the βCD and the 1,4-butanediol molecules, ring puckering parameters[12] for the glucose subunits, hydrogen-bonding distances for $d_{O \cdots O} < 3.5$ Å, and the measured and calculated structure factor amplitudes have been deposited with, and can be obtained from, Elsevier Science Publishers BV, BBA Data Deposition, P.O. Box 1527, Amsterdam, The Netherlands. Reference should be made to No. BBA/DD/492/*Carbohydr. Res.*, 228 (1992) 321–332.

The positions of the five interstitial water molecules are similar to those in the isomorphous complexes *β*CD·12H$_2$O[4] and *β*CD·EtOH·8H$_2$O[3,5] (W-4 is labelled as W-7 in these structures), and even the two-fold disorder of W-3 is observed in these complexes. In *β*CD·12H$_2$O, W-5 is also two-fold disordered. The positions of the interstitial water molecules are determined by the packing of the *β*CD molecules and are independent of the guest molecule.

The βCD molecule. — The *β*CD molecule is in the same conformation as in the isomorphous crystal structures, and the geometry has been described in detail[4,14]. The only substantial variation in the conformation in these structures is the orientation of the C-6–O-6 groups, which may point "away" from the molecular axis with the torsion angles O-5–C-5–C-6–O-6 of ~ −60°, or point "inside" with a torsion angle of ~ +60°. In many instances, disorder between these conformations has been observed, but none was found in the present structure. The bond C-6^4–O-6^4 points "inside" to form a hydrogen bond to a water molecule enclosed in the cavity, and all other C-6–O-6 bonds

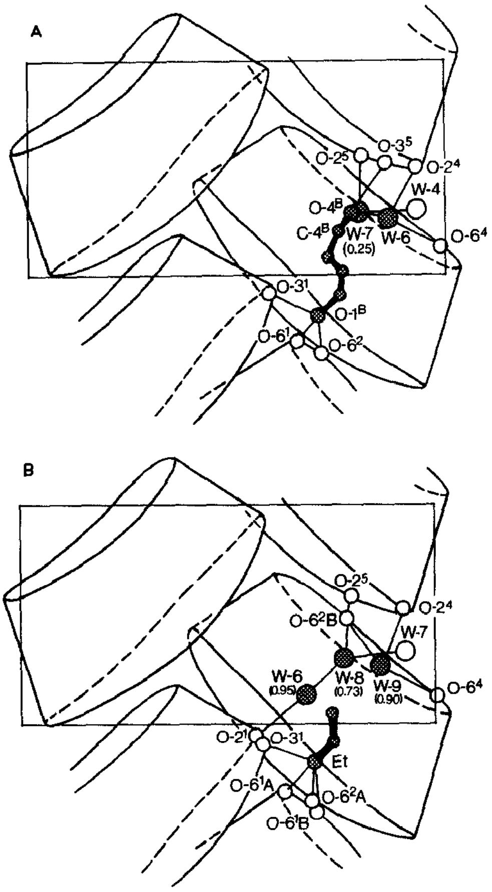

Fig. 2. Hydrogen-bonding arrangement of the molecules enclosed in the cavity in (**A**) *β*CD–1,4-butanediol·6.25H$_2$O and (**B**) *β*CD–ethanol·8H$_2$O (room temperature, X-ray study[3]), occupancy factors in parentheses.

point away from the molecular axis. A more detailed comparison of these orientations in the isomorphous structure has been given[3].

The guest molecules. — The 1,4-butanediol molecule is arranged roughly along the molecular axis of the βCD. The apolar $(CH_2)_4$ moiety occupies the central region of the cavity, which has a rather hydrophobic wall, and the hydroxyl groups are placed at the openings and engaged in strong hydrogen bonds with adjacent molecules (Fig. 2). The 1.25 included water molecules are situated at the narrow end of the cavity, roughly in the C-6 plane. The 1,4-butanediol hydroxyl group HO-1^B, which is located at the wider end of the cavity in the O-2/O-3 plane, forms three hydrogen bonds with hydroxyl groups of two neighboring βCD molecules. At the other opening of the cavity, HO-4^B forms hydrogen bonds with the HO-3^5 of a neighboring βCD molecule and with the included water molecules W-6 and (if present) W-7. W-6 is hydrogen-bonded to HO-4^B, to HO-6^4 of the enclosing βCD molecule, and to HO-2^4 of a neighboring βCD molecule. W-7, if present, is hydrogen-bonded to HO-4^B, to HO-2^5 of a neighboring βCD molecule, and to W-4, which is located "above" the O-6-rim of the enclosing macrocycle.

The distance W-7···C-4^B is only 2.18 Å and too short for a van der Waals contact; that of W-7···O-4^B, 2.61 Å, is also short for a hydrogen bond in a carbohydrate structure. This finding implies that W-7, if present, pushes C-4^B and, to a smaller extent, O-4^B, from the positions which they occupy if W-7 is not present. In crystallographic refinement, such an effect is difficult to resolve, since the occupancies of W-7, and hence of the alternative sites of C-4^B and O-4^B are only ~ 0.25, and the distances between the alternative sites will be shorter than the limit of crystallographic resolution (~ 0.9 Å). Consequently, it was not possible to assign the positions of these atoms to two sites with occupancies of ~ 0.75 and 0.25, which, in fact, are indicated by the somewhat irregularly shaped electron-density distributions of O-4^B and C-4^B (Fig. 3). Consequently, the

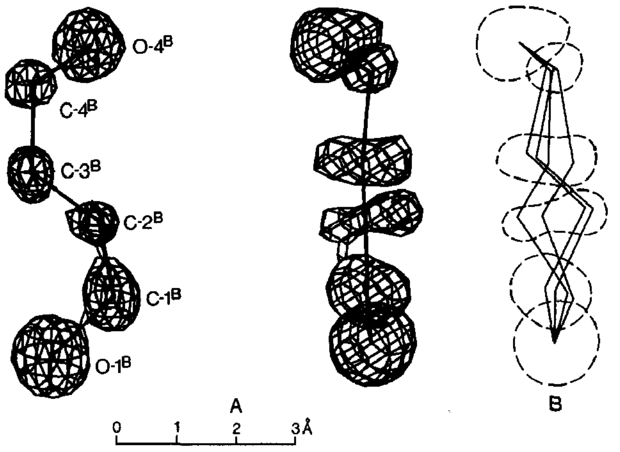

Fig. 3. (A) Electron-density distribution of the 1,4-butanediol molecule in two projections mutually perpendicular, displayed by FRODO[8,9] at the 1 x r.m.s.-density contour level. (B) Four model conformations that fit into the electron density distribution, based on the simplifying assumptions that the oxygen atoms are fixed and that the water position W-7 is not occupied.

positions of C-4^B and O-4^B given in the final set of co-ordinates are the weighted means of the alternative sites. Because of the greater occupancy of the major site, they will be close to the positions which the atoms occupy if W-7 is not present in the βCD cavity.

The $(CH_2)_4$ moiety of the 1,4-butanediol molecule occupies the largest part of the βCD cavity. The rather "thin" chain cannot completely fill the cavity, which has a diameter of ~ 7 Å and can accommodate much bulkier guest molecules (*e.g.*, hexamethylenetetramine[16]). This situation implies that the chain has a considerable scope for thermal motions.

The arrangement of the guest molecules in some aspects parallels that in βCD–ethanol octahydrate[3], where the solvent molecules are markedly disordered. In its major position, the ethanol, which can be regarded as "half" a 1,4-butanediol molecule, has its hydroxyl group hydrogen-bonded in the same position as O-1^B (Fig. 2), and the CH_2CH_3 moiety is oriented into the cavity and vibrates extensively. W-8 and W-9 are in almost the same positions as those of W-7 and W-6, respectively, in the present structure, and form a similar hydrogen-bonding arrangement. W-6 (ethanol complex), however, which is located in the central part of the cavity, is displaced by the 1,4-butanediol molecule, and, in the ethanol complex, no atom occupies the place equivalent to that of O-4^B.

The 1,4-butanediol molecule. — The electron density distribution of the 1,4-butanediol molecule, as obtained by $2F_o - F_c$ Fourier synthesis from the final set of co-ordinates, is shown in Fig. 3. At the contour level displayed, the oxygens appear to be larger than the carbon atoms, since they contain more electrons. C-1^B appears to be larger than the other carbons, as it is better ordered as shown by the temperature factors [$U_{eq.}$ values: O-1^B 0.046(6), O-4^B and C-1^B 0.09(1), C-2^B 0.22(2), C-3^B 0.16(2), and C-4^B 0.15(2) Å^2]. The electron density for O-1^B has an almost spherical distribution at all contour levels, and those of C-1^B, C-4^B, and O-4^B are spherical with some irregularities. The distribution of the electron densities for C-2^B and C-3^B is elongated and the former has a slight dumb-bell appearance, the distance of the dumb-bell "maxima" being > 1 Å. This finding is indicative of disorder or extensive thermal vibration with some preference for the outer positions. Attempts to describe these positions as two individual sites in the refinement were unsuccessful, and only the mean positions are given in the set of atomic co-ordinates. According to the distribution of the electron density, the "true" positions of C-2^B and C-3^B may at any time deviate by ~ 0.5 Å from these mean positions.

If only the mean atom positions are regarded, the covalent geometry of the refined molecule is poor. Except for O-1^B–C-1^B, the bond distances are too short and the bond angles are too large; the extreme cases are C-1^B–C-2$^B_{mean}$ (1.30 Å) and C-1^B–C-2$^B_{mean}$ (136°). Furthermore, the torsion angle O-1^B–C-1^B–C-2$^B_{mean}$–C-3$^B_{mean}$ [5(4)°] is in the energetically disadvantageous *cis*-range, and should tend to flip to the (+)-*gauche* or the (–)-*gauche* range. It is assumed that the observed mean positions do *not* represent an equilibrium conformation, around which some thermal vibrations are performed, but an *apparent* conformation that can be described as the superposition of two or more energetically allowed conformations.

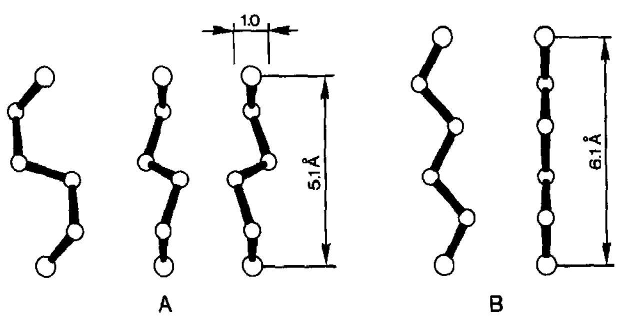

Fig. 4. (A) (*left*) Schematic view of 1,4-butanediol in the ideal low-energy conformation with torsion angles (±)-*gauche*, *trans*, (±)-*gauche*, $d_{O\cdots O}$ = 5.1 Å. The S-shaped side-view is similar to that of the experimental electron density. The *superposition* of the two possible conformations (*center* and *right*) resembles the experimental electron density. In refinement, a model of two superimposed ideal conformations proved to be too simple. (B) The elongated zigzag conformation (*trans*, *trans*, *trans*) of 1,4-butanediol with $d_{O\cdots O}$ = 6.1 Å.

Thus, the electron-density distribution may be described as the superposition of two half-occupied molecules with the conformations shown in Fig. 4A in an ideal form with the torsion angles O-1–C-1–C-2–C-3 and C-2–C-3–C-4–O-4 (±)-*gauche* and C-1–C-2–C-3–C-4 *trans*. The "side-view" of the molecule is similarly S-shaped, as observed experimentally (Fig. 3A). If O-1, C-1, C-4, and O-4 of these two conformations have the same location, then the distances between the alternative sites of C-2 and C-3 are ~ 1.0 Å each. This value corresponds to the distance between the above described "dumb-bell" maxima; the O···O distance is 5.12 Å, almost the same as that (5.15 Å) found experimentally.

Attempts to refine the molecule as the mean of only two partially occupied sites constrained close to ideal geometry were unsatisfactory, probably due to the limited crystallographic resolution, or to a more complex situation in the crystal. It is possible to draw numerous conformations of 1,4-butanediol with good covalent geometry into the electron-density distribution, as shown by the four examples in Fig. 3B. The situation is further complicated by the positional disorder of O-4^B and C-4^B imposed by the absence or presence of the water molecule W-7. Thus, a superpostion of only two alternative conformations is too simple a model to describe the 1,4-butanediol molecule appropriately.

At room temperature, the molecule is flexible and may adopt several conformations. The four examples shown in Fig. 3B are based on the assumption that the positions of the oxygen atoms are fixed. Actually, the oxygen atoms also perform thermal motions and, although smaller than those of the carbon atoms, these motions will increase the variety of conformations that fit into the crystallographic electron density. Consequently, the motion of the flexible chain of 1,4-butanediol is complex, although it is confined to the regions defined by the experimentally observed electron density. These motions *cannot* be compared with those of a vibrating chord, because the *mean positions* of the moving atoms do not represent an equilibrium conformation, as outlined above.

The conformations of low energy, which have intramolecular torsion angles in the (±)-*gauche* and the *trans* ranges, have discrete O···O distances. The planar zigzag conformation (torsion angles *trans*, *trans*, *trans*) has an O···O distance $d_{O\cdots O}$ of 6.1 Å (Fig. 4B). For the next-shorter low-energy conformation (*trans*, ±-*gauche*, *trans*), $d_{O\cdots O}$ is ~ 5.6 Å, followed by $d_{O\cdots O}$ ~ 5.1 Å (*trans*, ±-*gauche*, *trans* and ±-*gauche*, *trans*, ±-*gauche*), 4.6 Å (±-*gauche*, *trans*, ±-*gauche*), and so on. Thus, only for certain O···O distances may the $(CH_2)_4$ chain adopt a stereochemically favourable conformation. For the 1,4-butanediol molecule enclosed in the βCD cavity, the positions of the hydroxyl groups will be determined by two different (and, in the general case, competing) contributions to the potential energy. The hydroxyl groups will try to occupy positions of optimum hydrogen-bonding potential, whereas the $(CH_2)_4$ chain demands one of the discrete ideal O···O distances given above (5.15 Å in the present example).

The hydrogen-bonding network. — The crystal structure contains a large number of O–H···O hydrogen bonds, which are interconnected to form a spatial network. The hydrogen-bonding arrangement around the βCD molecule is shown in Fig. 5A, and for the βCD cavity in Fig. 5B (for O···O distances <3.2 Å). Due to the isomorphous

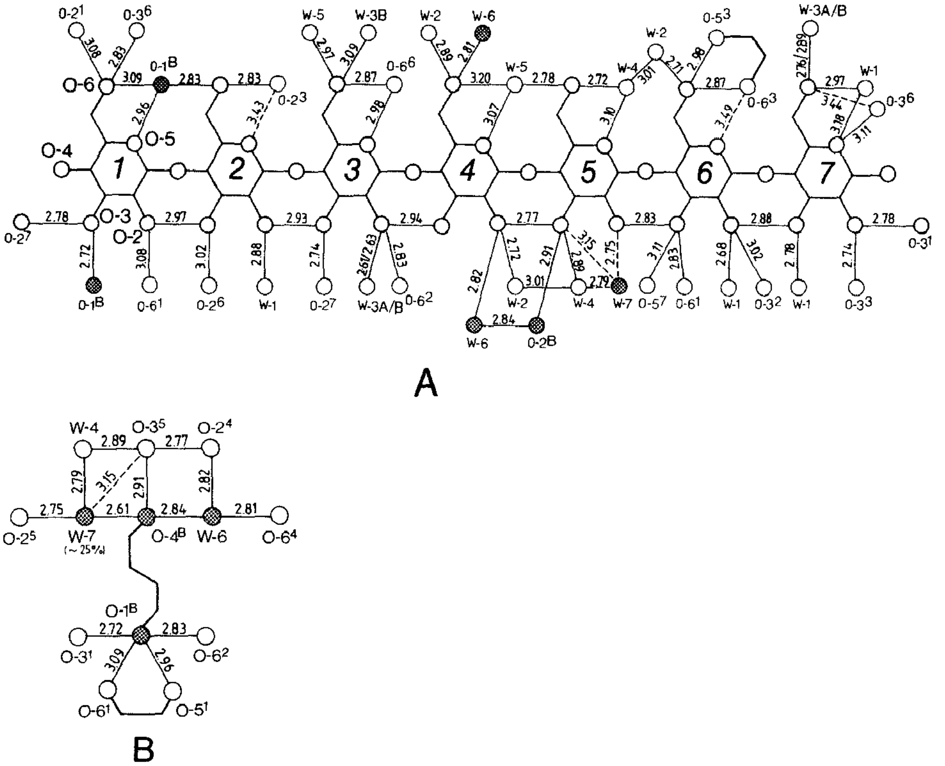

Fig. 5. Schematic representation of the hydrogen-bonding arrangements around (A) the βCD and (B) the 1,4-butanediol molecules ($d_{O\cdots O}$ <3.2 Å). Shaded atoms are enclosed in the βCD cavity. Intramolecular distances O-2···O-4, O-3···O-4, and O-5···O-6 are always <3.2 Å; for stereochemical reasons, they may represent only minor components of three-center hydrogen bonds.

packing in the crystal, the hydrogen-bonding arrangement parallels in many details that in βCD–ethanol octahydrate[3]. The ring of strong intramolecular inter-glucose O-2···O-3 hydrogen bonds is a characteristic of the βCD molecule and has been found in all crystal structures published so far. As most hydrogen atoms could not be located unambigously, the orientations of the hydrogen bonds cannot be given. In particular, it cannot be decided whether the hydrogen bonds are disordered orientationally, as found in the neutron-determined crystal structures of βCD undecahydrate[17] and of βCD–ethanol octahydrate[3] (dynamical "*flip-flop*" disorder), or ordered as reported for the isomorphous complex βCD–benzyl alcohol pentahydrate[14] (X-ray crystal structure).

A striking detail in the hydrogen-bonding network is the system of three interconnected, four-membered hydrogen-bonding rings (Fig. 6). The three rings are roughly square with O···O distances between 2.72 and 3.01 Å and O···O···O co-ordination angles between 84.1 and 92.3°. As the ideal co-ordination geometry for O–H···O hydrogen bonds is tetrahedral with co-ordination angles of ~ 110°, such square arrangements represent substantial distortions from ideal geometry. Single four-membered hydrogen-bonding rings have been found in many cyclodextrin crystal structures, for example, in βCD undecahydrate (neutron structure[17]) and γCD hexadecahydrate (neutron structure[18]), and a system of two such rings is observed in βCD–ethanol octahydrate (neutron structure[3]). However, the existence of a system consisting of three four-membered hydrogen-bond rings is surprising. As the dihedral angles between the least-squares planes of the rings (97.5°, 82.1°, and 102.0°) deviate significantly from 90°, the arrangement as a whole should not be regarded as "cubic". If the weakly populated water site W-7 is occupied, even a fourth four-membered ring (W-7–O-4^B–O-3^5–W-4–W-7) joins the system. As W-7 and O-4^B are not well-refined due to disorder, the geometry of that ring is much less accurately defined than the others.

Chelation of hydroxyl groups and water molecules between O-6 and O-5 of the glucose residues. — In cyclodextrin crystal structures, water molecules and hydroxyl groups are frequently found chelated between O-6 and O-5 at the primary hydroxyl rim. The chelated groups often also form hydrogen-bond bridges between HO-6 groups of

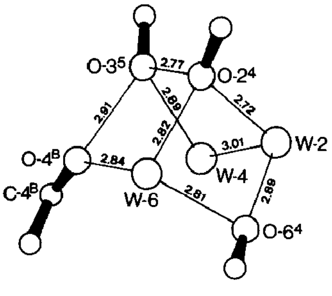

Fig. 6. System of three four-membered hydrogen-bonding rings of almost square geometry. The projection is as in Figs. 1 and 2; the O···O distances are given in Å. For the βCD hydroxyl groups, the C–O carbon atoms, and for HO-4^B of 1,4-butanediol two carbon atoms are drawn for clarity. In the isomorphous βCD–ethanol octahydrate (neutron crystal structure[3]), a system of two such hydrogen-bonding rings was found in the equivalent position (O-3^5–O-2^4–W-2–W-7–O-3^5 and O-2^4–W-2–O-6^4–W-9–O-2^4 (W-4 is labeled as W-7, and W-6 as W-9 in that report[3]).

adjacent glucose residues. Several of these arrangements are also observed in the present structure (Fig. 5A). A strong chelation was found for HO-1^{B} of the 1,4-butanediol molecule, which simultaneously forms contacts of <3.2 Å with O-5^{1}, O-6^{1}, and O-6^{2}.

The hydrogen-bonding arrangements cannot be described in detail from X-ray crystal structures of larger molecules, since the hydrogen positions are not determined with sufficient accuracy. Systematic studies of such chelated contacts in the six available neutron crystal structures of cyclodextrins, where the hydrogen atoms were located with high precision, showed[3] that these arrangements usually represent three-centre hydrogen bonds. Thus, either HO-6 donates hydrogen bonds to the chelated atom *and* to O-5, or the chelated atom donates hydrogen bonds to O-6 and to O-5. Simple two-centre hydrogen bonds are found rarely, and in no case was the arrangement O-6–H···O–H···O-5 observed. An analysis of the O···O distances showed that simultaneous contacts of a chelated atom to O-5 and O-6 with $d_{O\cdots O} < 3.5$ Å is indicative of a corresponding three-centre hydrogen bond with a reliability of ~ 90%. This is a powerful tool to identify such three-centre bonds in X-ray studies, where only the oxygen positions are determined.

On the basis of this study[3], chelating three-centre hydrogen bonds were identified at glucose residues 1, 3, 4, 5, and 7; the contacts at glucose residues 2 and 6 have rather long O···O-5 distances and may also be two-centre hydrogen bonds with van der Waals contact to O-5. Figs. 7 and 8 show two examples of such arrangements together with arrangements of similar geometry found in the neutron crystal structure of βCD–ethanol octahydrate[3]. The chelation of O-6^{6} between O-6^{3} and O-5^{3} is observed in almost identical geometry in the ethanol complex, where O-6^{6} acts as the donor of a three-centre hydrogen bond (Fig. 7). The chelation of O-1^{B} between O-6^{1}, O-5^{1}, and O-6^{2} and the additional hydrogen bond with O-3^{1} of a symmetry-related βCD molecule is found also for the ethanol molecule in βCD–ethanol octahydrate (Fig. 8). In this complex, HO-6^{1} was the donor of a three-centre hydrogen bond to O(Et) and O-5^{1}. Although the 1,4-butanediol and the ethanol molecules are engaged in virtually the same system of hydrogen bonds, the orientation of the hydroxyl groups, *i.e.*, the *directionality* of the hydrogen bonds may be different.

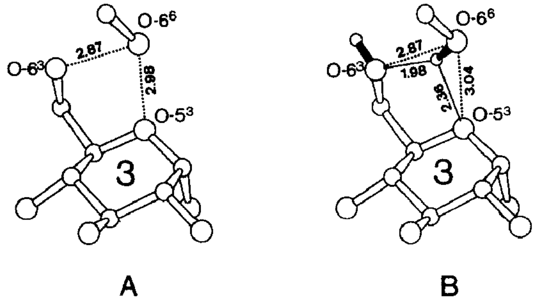

Fig. 7. Chelation of HO-6^{6} between O-6 and O-5 of glucose residue 3 in (A) the present structure and in (B) βCD–ethanol octahydrate, where a three-centre hydrogen bond is donated by the chelated group[3]. Note the similarity of the O···O distances.

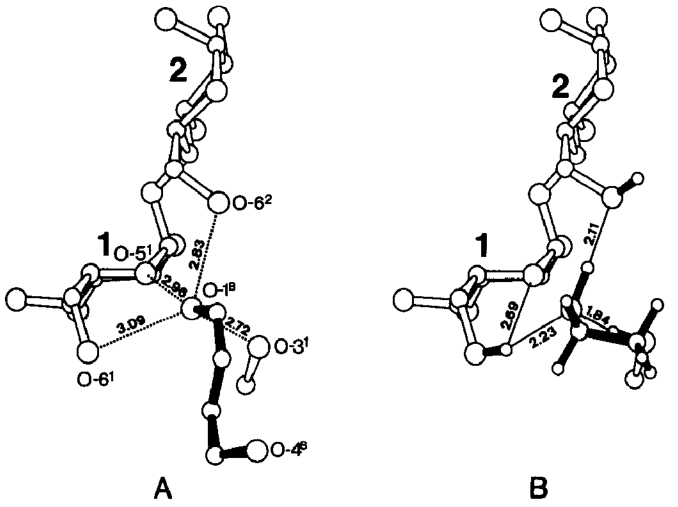

Fig. 8. (A) Chelation of a 1,4-butanediol hydroxyl group by O-6^1, O-5^1, and O-6^2, and an additional hydrogen bond to O-3^1 of a symmetry related molecule; and (B) a similar arrangement in βCD–ethanol octahydrate[3] (the structure is disordered in this region, and only the major atom sites are drawn). In B, HO-6^1 donates a three-centre hydrogen bond to O–Et and O-5^1; note the hydrogen-bond bridge O-6^1–H···O–Et–H···O-6^2. In A, the orientation of the hydrogen bonds may be different (in B, the O···O distances are $d_{O\text{-}6^1\cdots O\text{-}Et}$ = 3.07 Å, $d_{O\text{-}5^1\cdots O\text{-}Et}$ = 3.16 Å, $d_{O\text{-}Et\cdots O\text{-}6^2}$ = 3.16 Å).

Such chelated arrangements, which are also observed in the crystals of a linear hexasaccharide[19], seem to be a characteristic structural element of hydrated oligosaccharides in general and will be analysed[20] more systematically elsewhere.

Structure of a different individual crystal. — This study was initiated with the X-ray structure analysis of a crystal taken one week after crystallisation from the same crystallisation tube used for the experiment described above. Due to technical problems, data were collected only to a resolution of ~ 1.0 Å. As some disorder of the 1,4-butanediol molecule could not be resolved satisfactorily, the refinement was abandoned, although it converged at an R value of 0.061, and a new set of data was collected to higher resolution. The structure of this first crystal was practically identical with that reported in detail, except for the partially occupied water site W-7 and the atoms C-4^B and O-4^B of the 1,4-butanediol molecule. W-7 was in the same position, but had an occupancy factor ~ 0.5 instead of ~ 0.25. Consequently, the disorder of O-4^B and especially C-4^B, which have different positions depending on the presence or absence of W-7, was more substantial. The higher occupancy factor of W-7, which considerably complicated the refinement, again confirms the experience that, in crystal structures of this complexity, slight but significant differences may be observed between different individual crystals. Such delicate variations, which may be caused by slightly different conditions of crystallisation or by different histories after crystallisation, may be of importance if disorder phenomena are studied, as described in greater detail for βCD–ethanol octahydrate[3].

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