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Molecular dynamics simulations of β-cyclodextrin in aqueous solution

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Abstract

Molecular dynamics (MD) simulations of β-cyclodextrin (β-CD) have been carried out in aqueous solution at 300 K over a period of about 200 ps using Tripos force field. The atomic trajectories obtained by these simulations have been analysed by means of the occurrence of hydrogen bonds. The MD simulations lead to an association of seven water molecules into the β-CD cavity which is in a good agreement with X-ray crystallography experiments. This demonstrates that the force field used in the simulations is able to reproduce the experimentally observed hydrophilic–hydrophobic characteristics of β-CD molecule.

Keywords: β-Cyclodextrin; Molecular dynamics simulation; Hydrogen bonding; Water–cyclodextrin inclusion

1. Introduction

β-Cyclodextrin (β-CD) is a cyclic oligosaccharide consisting of seven D-glucose units linked by α (1–4) interglucose bonds (Szejtli, 1998). The crystal structures of the β-CD complexes exhibit a large number of O–H···O hydrogen bonds between the three hydroxyl groups and water molecules, which are interconnected to form complicated three-dimensional networks (Betzel et al., 1984). In neutron diffraction analysis, the C2-OH groups of the glucose units form flip-flop hydrogen bonds with the C3-OH groups of the adjacent glucose units. Therefore, β-CD has a rather rigid structure. This intramolecular hydrogen bond formation is also the explanation for its relatively low solubility in water compared to other CDs (Koehler et al., 1988). The ability of β-CD to form inclusion complexes with a variety of compounds is the basis for the use as pharmaceutical carrier molecules for drugs, which are unstable at ambient conditions or poorly water-soluble (Uekama et al., 1998). Since most of the reactions involving CDs take place in an aqueous environment, the interaction between CDs and water is of fundamental importance, as there exists a competition between hydrophobic guest molecules and the water molecules in the cavity bound by hydrogen bonding.

The structure of hydrated β-CD is well known from X-ray (β-CD·12H_2O; Linder and Saenger, 1982) as well as from neutron diffraction studies (β-CD·11H_2O; Zabel et al., 1986; Steiner and Koellner, 1994). About seven water molecules, in average, are located in the cavity and distributed over eight positions. To assess whether this also holds for β-CD dissolved in water and for testing the empirical force fields which are currently used in simulations

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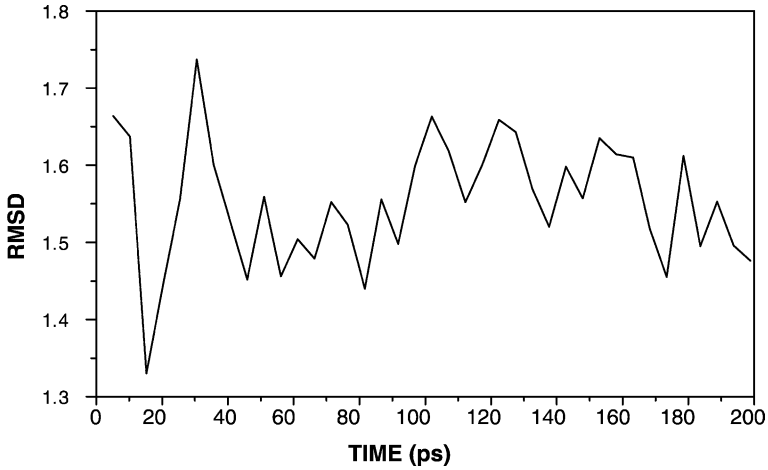

Fig. 1. Time dependence of the root mean square deviation (RMSD) of the β-CD heavy atoms along a typical MD trajectory.

of protein and nucleic acid structures, molecular dynamics (MD) simulations of this molecule in water solution were performed (Koehler et al., 1987, 1988; Melani et al., 1998; Braesicke et al., 2000; Momany and Willett, 2000; Winkler et al., 2000; Starikov et al., 2001).

The aim of the present study is to establish a molecular model able to simulate the behaviour of β-CD in aqueous solution by applying the empirical Tripos force field (Clark et al., 1989). We focused our attention on the water molecules distributed around β-CD as well as inside the cavity. Particularly, hydrogen bonding in the system was of central interest.

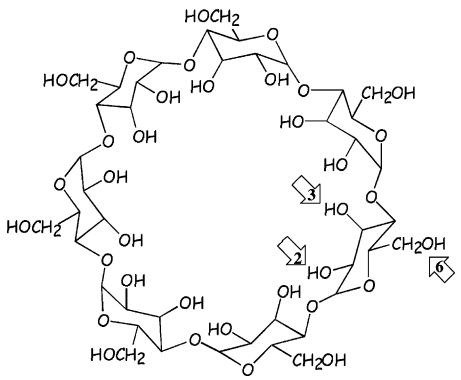

Fig. 2. Structure of the β-CD, the primary hydroxyl group (O6-H) and the secondary hydroxyl groups (O2-H, O3-H) located on the edge of β-CD ring.

Trajectories, which are simply a history of the motion of the system over a certain time period, were generated. The MD included both kinetic and potential energies obtained from empirical force field. Generally, the advantage of MD is that by applying enough kinetic energy (temperature) one can move around on a molecule's potential energy surface, sampling many conformational states along the motion path.

Table 1
The occurrence of hydrogen bonds in the MD simulations, listed according to the donor or acceptor function of the specified atom

Time (ps)	Donor	Acceptor						
		O2	O3	O4	O5	O6	OW	Sum
71.4	O2	0	1	1	0	0	3	5
112.2		0	1	1	0	0	2	4
188.7		0	1	1	0	0	3	5
71.4	O3	1	0	0	0	0	3	4
112.2		1	0	0	0	0	3	4
188.7		1	0	0	0	0	1	2
71.4	O6	0	0	1	2	0	2	5
112.2		0	1	1	2	0	2	6
188.7		0	0	2	2	0	3	7
71.4	OW	3	5	7	6	9	19	49
112.2		6	5	8	9	9	21	58
188.7		9	5	7	10	9	28	68
71.4	Sum	4	6	9	8	9	27	63
112.2		7	7	10	11	9	28	72
188.7		10	6	10	12	9	35	82

Table 2
Hydrogen bonds of β-CD at 300 K from the MD simulations

Donor		Acceptor	H···A (Å)			
			0 ps	71.4 ps	112.2 ps	188.7 ps
(a) Glucose → glucose						
O2(1)	H2	O3(2)	1.57	1.66	1.72	1.71
O6(3)	H10	O5(2)	1.59	1.56	1.72	1.70
O6(3)	H10	O4(3)	2.21	2.37	1.78	2.18
O3(4)	H19	O2(3)	–	–	–	2.42
O6(4)	H20	O5(3)	1.65	–	–	–
O2(5)	H28	O4(5)	1.77	1.83	1.78	1.56
O6(5)	H30	O3(5)	–	–	1.95	–
O6(6)	H40	O5(5)	1.59	1.53	1.61	2.11
O6(6)	H40	O4(6)	2.43	–	–	1.73
O3(7)	H49	O2(6)	–	1.71	2.06	–
(b) Glucose → water						
O3(1)	H3	OW8	–	1.54	1.71	–
O3(2)	H6	OW32	–	–	–	2.41
O6(2)	H7	OW26	–	1.76	1.80	1.93
O2(3)	H8	OW4	1.58	1.64	1.62	1.68
O3(3)	H9	OW45	–	2.39	–	–
O3(4)	H19	OW6	–	2.29	1.67	–
O6(4)	H20	OW25	–	2.01	2.01	1.98
O6(5)	H30	OW22	1.66	–	–	–
O2(6)	H38	OW19	–	2.34	–	2.35
O3(6)	H39	OW10	–	–	1.55	–
O3(6)	H39	OW20	2.47	–	–	–
O2(7)	H48	OW23	–	2.22	1.87	2.27
O2(7)	H48	OW30	1.87	–	–	–
O6(7)	H50	OW16	–	–	–	1.94
(c) Water → glucose						
OW38	HW2	O2(1)	–	–	–	1.96
OW13	HW1	O3(1)	–	–	–	1.63
OW13	HW2	O3(1)	–	1.60	1.72	–
OW7	HW1	O4(1)	1.54	1.53	1.59	1.43
OW7	HW2	O4(1)	2.24	2.43	2.22	2.14
OW2	HW1	O5(1)	1.65	2.07	2.17	1.86
OW2	HW2	O5(1)	1.91	1.88	1.61	–
OW27	HW2	O5(1)	–	–	–	1.50
OW5	HW2	O6(1)	–	1.60	1.64	1.56
OW32	HW2	O2(2)	–	–	–	1.64
OW2	HW1	O4(2)	1.57	1.54	1.78	1.98
OW2	HW2	O4(2)	–	–	2.47	–
OW45	HW1	O5(2)	–	–	2.43	–
OW45	HW2	O5(2)	–	–	1.54	1.68
OW45	HW1	O6(2)	–	2.37	1.68	1.50
OW45	HW2	O6(2)	–	1.64	–	2.31
OW6	HW1	O2(3)	2.47	1.53	1.59	1.57
OW6	HW2	O2(3)	1.59	–	–	–
OW41	HW1	O3(3)	1.58	1.62	1.65	1.68
OW42	HW1	O4(3)	1.50	–	–	–
OW42	HW2	O4(3)	–	1.50	1.48	1.51
OW25	HW2	O5(3)	–	2.36	2.47	2.01
OW31	HW2	O5(3)	–	–	1.73	–

Table 2 (*Continued*)

Donor		Acceptor	H···A (Å)			
			0 ps	71.4 ps	112.2 ps	188.7 ps
OW34	HW2	O5(3)	–	1.63	1.59	1.60
OW23	HW2	O6(3)	–	1.63	1.59	1.62
OW42	HW1	O6(3)	2.30	–	–	–
OW42	HW2	O6(3)	1.60	–	–	–
OW43	HW1	O2(4)	–	–	2.38	1.69
OW6	HW1	O3(4)	2.38	–	–	–
OW44	HW1	O3(4)	1.60	1.65	1.66	1.57
OW6	HW1	O4(4)	1.58	–	–	–
OW25	HW2	O4(4)	–	1.71	1.81	1.78
OW39	HW1	O5(4)	–	–	–	1.48
OW3	HW2	O6(4)	1.58	1.53	1.54	1.61
OW29	HW2	O6(4)	–	–	–	2.41
OW30	HW1	O2(5)	1.64	–	–	1.61
OW30	HW2	O2(5)	–	–	2.43	2.31
OW43	HW1	O2(5)	–	1.53	1.70	1.74
OW22	HW2	O5(5)	–	–	–	1.61
OW35	HW1	O6(5)	–	1.62	1.74	1.72
OW35	HW2	O6(5)	–	2.41	–	–
OW10	HW2	O2(6)	–	–	1.59	2.09
OW47	HW1	O2(6)	1.56	–	–	–
OW20	HW1	O3(6)	–	1.50	1.62	1.94
OW46	HW1	O3(6)	1.64	–	–	–
OW30	HW2	O4(6)	1.54	1.57	1.66	1.61
OW19	HW2	O5(6)	1.59	–	–	–
OW25	HW1	O6(6)	–	1.59	1.65	1.60
OW25	HW2	O6(6)	–	–	2.43	–
OW14	HW1	O2(7)	1.56	1.56	1.53	1.52
OW28	HW1	O2(7)	2.34	–	–	–
OW33	HW2	O3(7)	–	1.53	1.63	1.64
OW19	HW1	O4(7)	–	1.77	–	–
OW19	HW2	O4(7)	–	–	1.52	1.56
OW7	HW1	O5(7)	2.19	2.16	1.86	2.28
OW7	HW2	O5(7)	1.54	1.53	1.59	1.66
OW16	HW1	O5(7)	–	–	–	1.71
OW1	HW1	O6(7)	–	–	2.03	–
OW9	HW1	O6(7)	1.58	–	1.48	1.65
OW9	HW2	O6(7)	–	1.53	–	–
(d) Water → water						
OW1	HW1	OW24	–	–	1.48	–
OW1	HW2	OW24	–	–	2.28	–
OW4	HW1	OW3	1.55	1.53	1.75	–
OW4	HW2	OW3	-	–	–	1.52
OW4	HW2	OW36	–	1.55	–	–
OW5	HW1	OW13	–	1.69	1.63	1.74
OW8	HW1	OW14	–	–	1.66	–
OW8	HW2	OW14	–	1.67	–	–
OW9	HW1	OW1	–	–	2.36	–
OW9	HW2	OW1	–	–	1.89	–
OW11	HW1	OW26	–	1.59	–	–
OW11	HW2	OW2	1.54	1.76	–	–
OW12	HW1	OW29	–	–	–	1.71

Table 2 (*Continued*)

Donor		Acceptor	H···A (Å)			
			0 ps	71.4 ps	112.2 ps	188.7 ps
OW13	HW1	OW33	–	1.52	1.56	–
OW13	HW2	OW33	–	–	–	1.60
OW14	HW1	OW42	2.39	–	–	–
OW14	HW2	OW42	1.64	1.64	1.60	1.56
OW15	HW2	OW19	–	–	–	1.76
OW16	HW2	OW5	-	–	–	1.61
OW18	HW1	OW26	–	–	–	1.51
OW19	HW1	OW9	1.55	–	1.50	1.53
OW19	HW2	OW9	–	1.65	–	–
OW20	HW1	OW47	1.54	–	–	–
OW20	HW2	OW47	–	1.66	1.62	1.66
OW22	HW1	OW46	1.54	–	–	–
OW26	HW1	OW23	–	–	–	2.40
OW26	HW2	OW23	–	1.58	–	1.53
OW23	HW1	OW7	–	1.78	–	1.66
OW24	HW1	OW16	–	–	–	1.54
OW26	HW1	OW27	–	–	–	1.61
OW28	HW1	OW30	1.67	–	–	–
OW30	HW1	OW14	–	1.69	1.67	–
OW31	HW1	OW25	–	–	1.89	2.32
OW31	HW2	OW25	–	–	1.69	1.50
OW32	HW1	OW21	–	–	–	1.51
OW34	HW1	OW31	–	–	1.92	1.71
OW34	HW2	OW31	–	–	2.47	–
OW35	HW2	OW12	–	1.48	–	1.59
OW36	HW2	OW34	–	1.71	–	–
OW37	HW1	OW32	–	–	–	1.58
OW38	HW1	OW37	–	–	–	1.60
OW38	HW2	OW37	–	–	–	2.09
OW39	HW1	OW35	–	–	–	2.32
OW39	HW2	OW35	–	–	–	1.57
OW40	HW1	OW17	–	–	1.65	–
OW40	HW2	OW45	–	–	1.58	–
OW41	HW1	OW6	–	2.33	–	–
OW41	HW2	OW6	–	1.58	1.65	1.53
OW42	HW1	OW44	–	1.62	1.58	1.65
OW43	HW2	OW44	–	1.66	1.89	–
OW46	HW2	OW20	1.53	–	–	–
OW47	HW1	OW15	–	–	–	1.51

2. Method of calculations

The initial configuration of β-CD has been taken from Cambridge crystallographic data base (BCDEXD10; Linder and Saenger, 1982). The partial atomic charges of the molecule were obtained from ab initio calculations (HF/6-31G(d,p)) using Gaussian 98 program (Frisch et al., 1998). To investigate the conformational flexibility, we generated a unit cell containing one β-CD molecule in a water box with periodic boundary conditions, employing the Tripos force field (Clark et al., 1989) implemented in the Sybyl 6.7 program (Tripos Associates Inc., 2000) running on a Silicon Graphic workstation. The structures of 827 water molecules with β-CD fixed in its initial conformation were optimised (steepest descent method). An initial MD run was performed, for period of 10 ps at 300 K. This solvent equilibration

phase should be sufficiently extensive to allow the solvent to readjust completely to the potential field of the solute. The cut-off for non-bonded interactions was taken to be 12 Å throughout all the simulations.

At the beginning, we carried out high temperature annealed MD simulations starting at 1000 K (2 ps) annealing to 0 K (10 ps). The temperature of 1000 K is necessary to enable the molecule to overcome energy barriers between different conformations and to prevent the system from getting stuck in a particular region of the conformational space. Simulations at lower temperatures yielded in very similar conformations while at higher temperatures some distorted geometries of β-CD were obtained. The simulations of β-CD in aqueous solution were relaxed using the steepest descent method until a gradient different of 0.01 kcal/mol was reached. After energy minimisation of the system at 0 K, the MD simulation was initialised using a time step of 1 fs for a time period of 200 ps. The temperature was kept constant at 300 K yielding a canonical ensemble (NVT). To demonstrate the quality of the simulation data, the root mean square deviations (RMSD) of the simulated molecular systems are depicted by averaging over time intervals of 5 ps (40 data points) (Fig. 1). The average structures are determined from the minimum value of the |RMSD−Mean(RMSD)|. The RMSD values were obtained from superposing of the heavy atoms β-CD at each time with the structure at the beginning of the run (the structure after simulated annealing). The average all data points (Mean(RMSD)) was 1.533. Three of the average simulation structures were taken at the simulation times 71.4, 112.2 and 188.7 ps with |RMSD−Mean(RMSD)| difference values 0.001, 0 and 0.001, respectively. In the analysis of the MD data, as criterion for the existence of a hydrogen bond between donor (D) and acceptor (A), a distance D–H $\cdots$ A $<$ 2.5 Å was used (Koehler et al., 1988).

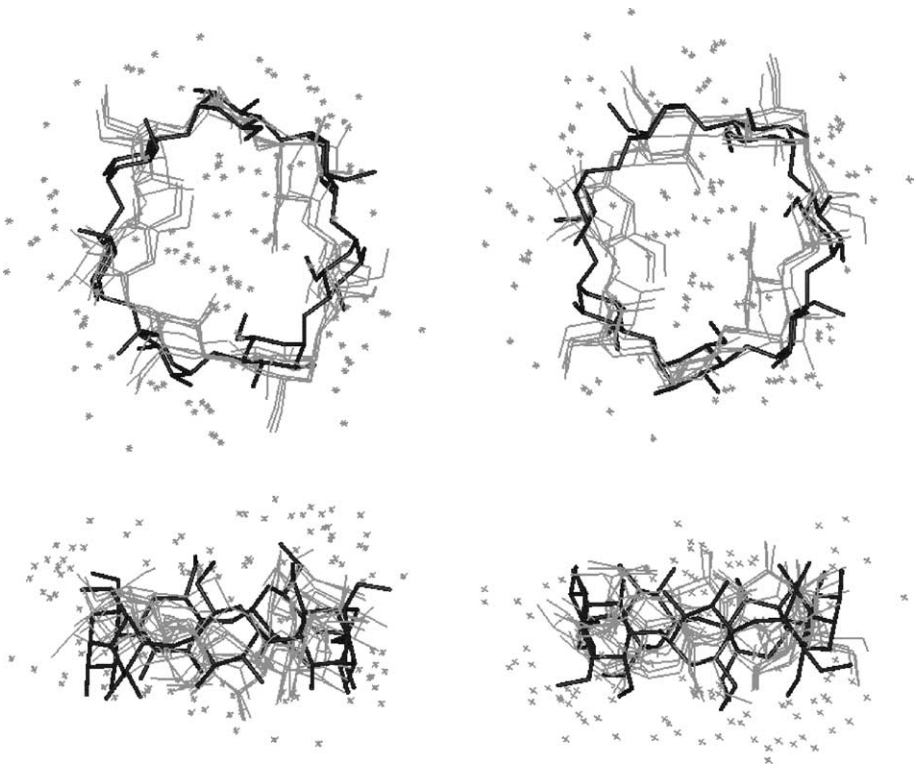

Fig. 3. Structure of the β-CD and water molecules in the crystal structure (black stick) and MD (grey lines) seen from the O6 rim (top, left side), from the O2, O3 rim (top, right side), in a side view of O6 rim up (bottom, left side) and in a side view of O6 rim down (bottom, right side); hydrogen atoms are omitted for clarity.

3. Results and discussion

As each glucose unit contains one primary O6-H and two secondary O2-H, O3-H hydroxyl groups (Fig. 2), which can act simultaneously as hydrogen donors and as acceptors, and as water molecule are able to build up hydrogen bond networks with two hydroxyl groups, the structures of β-CD in water solution in general display O–H···O hydrogen bonds as shown in Tables 1 and 2. These data allow us to perform an analysis of the number of hydrogen bonds in the simulations. Table 1 displays the results for all possible combinations of the four donor and six acceptor atoms. Interglucose, intramolecular O2···H···O3 hydrogen bonds are formed at every simulation. Hydrogen bonds with O2 atoms as donor (O3 as acceptor) occur as frequently as hydrogen bonds with an O3 atom as donor (O2 as acceptor). Therefore, no clear distinction between O2 or O3 hydroxyl groups acting as hydrogen bond donor can be made. The O6 atoms are slightly better acceptors than the O2 and O3 atoms. The glucosidic O4 atoms act as hydrogen bond acceptors, and, therefore, hydrogen bonds with the O2 and O6 hydroxyl groups as hydrogen bonds donor are built. The glucosidic O5 atoms are hydrogen acceptors for hydrogen bonds with the O6 hydroxyl groups only.

In the MD simulations, the water molecules outside the cavity are randomly distributed in some locations, whereas inside the cavity they remain in a similar position. The average structures of β-CD in water solvent obtained from the simulations are shown in Fig. 3, together with the crystal structure of β-CD dodecahydrate determined by the X-ray experiment (Linder and Saenger, 1982). Only water molecules within a distance of 2.5 Å from the β-CD molecule in each trajectories are concerned. In the initial time period of simulation, the β-CD hydrate system has only 18 water molecules in the distance 2.5 Å from the β-CD; it is increasing to be 30, 31 and up to 39 water molecules at simulation times 71.4, 112.2 and 188.7 ps, respectively. The influence of the transport process of water molecules along the main pathway in the MD simulations is the consequence of hydrogen bonding of the water molecules themselves (OW–H···OW) and OW acting as hydrogen bond donors to hydrogen bond acceptors: O2 and O5; this is supported by data in Tables 1 and 2 (OW stands for a water molecule).

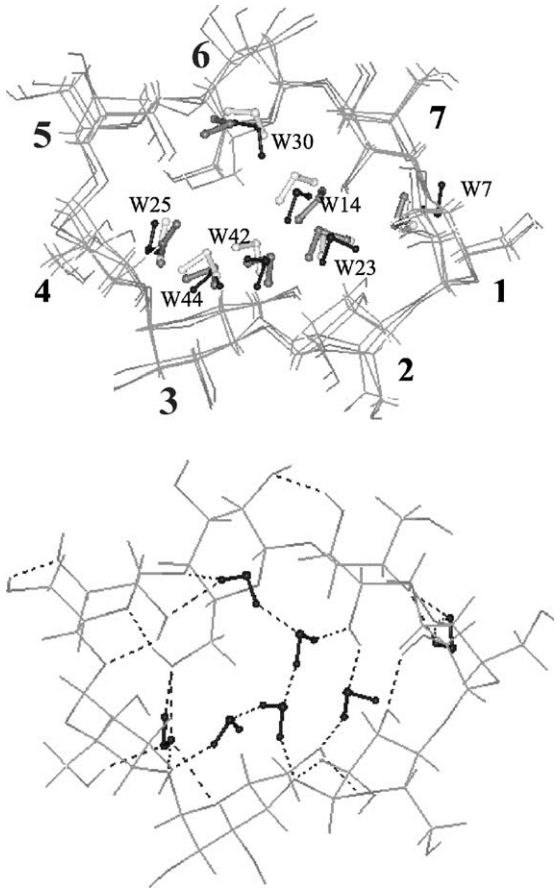

Fig. 4. Structure of the β-CD molecule hydrate in MD simulations, viewing onto the plane of the O4 atoms. Water molecules included in the cavity are also drawn in ball and stick models (light grey: 71.4 ps; black: 112.2 ps; dark grey: 118.7 ps; dashed lines: hydrogen bonds). Glucose residue numbering shown in italics. Inset shows the numbers of water molecules used in MD simulations.

From the results of the MD simulation, it can be clearly seen that the internal cavity of the β-CD molecule is occupied by water molecules. The number of these water molecules has been analysed in more detail to gain information of the binding positions and binding forces with β-CD. Fig. 4 shows the structure of β-CD hydrate in the MD simulations in detail. The projection is perpendicular to the equatorial plane of the β-CD molecule. The hydroxyl group are placed at the rims of the β-CD molecule, while the cavity surface is formed by hydrogen atoms bound to carbon atoms and by glucosidic oxygen atoms. Seven water molecules are enclosed in the cavity (W7, W14,

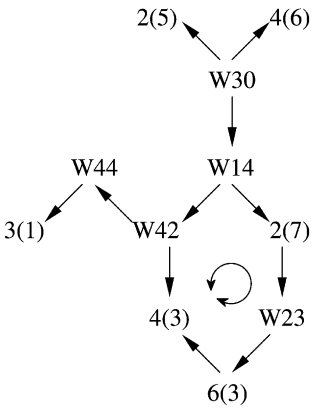

Fig. 5. The hydrogen bonds of the β-CD with the water molecules in the cavity, which consisted of cyclic 6-membered hydrogen bonded structure counter-running chain (antidromic).

W23, W25, W30, W42 and W44). They are distributed over eight positions as presented in Table 2 (b and c). Glucose-to-glucose, glucose-to-water and water-to-glucose hydrogen bonds of the average structure are also presented in Fig. 4. There are O–H · · · O hydrogen bonds engaged in a typical arrangement as collected in Fig. 5. These MD simulations of β-CD hydrate reveal a cyclic 6-membered hydrogen bonded structure (Fig. 5), which consisted of a counter-running chain, which is thus called antidromic (Saenger, 1979; Betzel et al., 1984; Zabel et al., 1986).

4. Conclusion

The results of the study agree with the generally accepted idea that CDs are members of a family of molecules with a hydrophilic character of the outer side, and hydrophobic character of the cavity. Although for water molecules, the interior of β-CD is more hydrophobic than the solvation shell in pure water, where extended hydrogen bond networks are present. Nevertheless, hydrogen bonds in a more rigid arrangement lead to the inclusion of seven water molecules. The β-CD hydrate structures obtained from MD simulations may serve as interesting model systems to investigate how conformational flexibility and dynamic disorder can influence inclusion complexes of a variety of compounds with β-CD. Moreover, the results represented in this paper demonstrate that the Tripos force field is able to reproduce the experimentally observed the hydrophilic–hydrophobic characteristics as well as hydrogen bonding of β-CD molecule in water, which is documented by the good agreement between X-ray crystallographic geometries compared to the MD simulations.

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