

## Hydrogen Bonding Analysis of Hydroxyl Groups in Glucose Aqueous Solutions by a Molecular Dynamics Simulation Study

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Molecular dynamics simulations have been performed to investigate hydrogen bonding characteristics of hydroxyl groups in glucose aqueous solutions with different concentrations. The hydrogen bonding abilities and strength of different O and H atom types have been calculated and compared. The acceptor/donor efficiencies have been predicted and it has been found that: (1) O2-HO2 and O3-HO3 are more efficient intramolecular hydrogen bonding acceptors than donors; (2) O1-HO1, O4-HO4 and O6-HO6 are more efficient intramolecular hydrogen bonding donors than acceptors; (5) O1-HO1 and O6-HO6 are more efficient intermolecular hydrogen bonding acceptors than donors while hydroxyl groups O2-HO2 and O4-HO4 are more efficient intermolecular hydrogen bonding donors than acceptors. The hydrogen bonding abilities of hydroxyl groups revealed that: (1) the hydrogen bonding ability of OH2-H<sub>w</sub> is larger than that of hydroxyl groups in glucose; (2) among the hydroxyl groups in glucose, the hydrogen bonding ability of O6-HO6 is the largest and the hydrogen bonding ability of O4-HO4 is the smallest; (3) the intermolecular hydrogen bonding ability of O6-HO6 is the largest; (4) the order for intramolecular hydrogen bonding abilities (from large to small) is O2-HO2, O1-HO1, O3-HO3, O6-HO6 and O4-HO4.

**Key Words :** Glucose, Hydrogen bond, Molecular dynamics simulation, Hydroxyl group

### Introduction

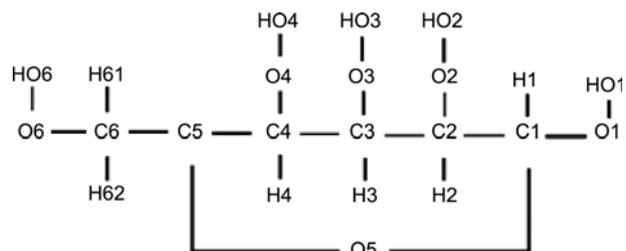
Intracellular ice formation (IIF) is an important physico-chemical parameter and it has a strong correlation with cellular death.<sup>1</sup> The cellular circumstance is composed of membrane, extra-cellular solutions, intra-cellular solutions with cryoprotective agents (CPAs), salt ions and so on. The structure and kinetics of CPA solutions play an important role in understanding the mechanisms of intracellular ice formation.<sup>2</sup> Glucose, also known as grape sugar, is a very important carbohydrate in biology and it can be used as a CPA in industry.<sup>3-6</sup> Thus, it's undoubtedly necessary to investigate the structure and kinetics of glucose aqueous solutions to explore its cryoprotective behavior as a CPA.

Many studies revealed that the structure and kinetics of glucose aqueous solutions were affected due to strong hydrogen bonds,<sup>7-16</sup> including loss of the tetrahedral ordering typical of the bulk water,<sup>7,8</sup> two hydrates composition with the pentahydrate (C<sub>6</sub>H<sub>12</sub>O<sub>6</sub>·5H<sub>2</sub>O) and the dihydrate (C<sub>6</sub>H<sub>12</sub>O<sub>6</sub>·3H<sub>2</sub>O),<sup>13</sup> reduction in water translational and rotational mobility<sup>7</sup> and decrease of glucose translational diffusion coefficients.<sup>16</sup> Hydrogen bonding structure and kinetics of glucose aqueous solutions have been studied and it has been found that water and glucose clusters were formed by hydrogen bonds<sup>17</sup> and the hydrogen bond lifetime increased as glucose concentration increases.<sup>14</sup>

There are five hydroxyl groups in a glucose molecule and hydrogen bonding abilities of these hydroxyl groups may be different. In fact, molecular dynamics simulation studies found that the hydroxyl groups were more efficient hydro-

gen bond donors than acceptors.<sup>12</sup> The structure and symbols of D-glucose molecules have been illustrated in Figure 1. A first-principle molecular dynamics simulations further found that: the anomeric site (O1-HO1) was the best donor, but the poorest acceptor among the hydroxyl groups; O2-HO2 was a slightly poorer donor; O3-HO3 was a better acceptor; O4-HO4 was a poorer acceptor; O6-HO6 was the best acceptor and a better donor as well.<sup>11</sup> However, the hydrogen bonding strength of these hydroxyl groups, the reason for different hydrogen bonding abilities, the concentration dependence of hydrogen bonding abilities and strength, acceptor/donor efficiency of hydroxyl groups and *et al.* are open to questions. In the present study, hydrogen bonding abilities and strength of different O(H) types were calculated. And then the acceptor/donor efficiency of molecules and hydroxyl groups have been analyzed as well as the hydrogen bonding abilities of hydroxyl groups.

The rest of the paper is organized as follows. In Sec.



**Figure 1.** The structure and symbols of D-glucose molecules used in the present study. The symbols for O and H atoms in water molecules are OH2 and H<sub>w</sub>, respectively.

Methods, we have presented the computation details and hydrogen bonds definition. The results including hydrogen bonding structure, kinetics and acceptor/donor analysis for different hydroxyl groups have been summarized in Sec. Results and Discussions. Our conclusions and outlooks are presented in final section.

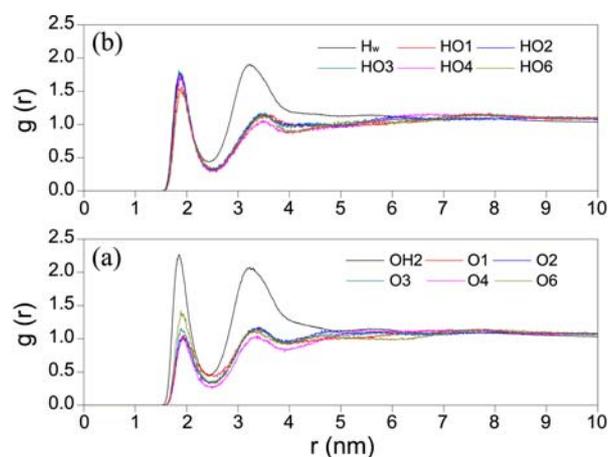
## Methods

### Computation Details.

**Molecular Dynamics Simulations:** In the present study, all simulations were done by molecular dynamics simulation package NAMD.<sup>18</sup> An additive empirical force field for hexopyranose monosaccharides with TIP3P water model was used to describe the interactions of glucose and water.<sup>19</sup> Periodic boundary conditions were used in three dimensions. The time step was 2 fs. Neighborhood lists were applied with a distance of 13.5 Å and lists were updated every 10 time steps. A switching function was used to truncate non-bonded interactions smoothly from 10.0 to 12.0 Å. Particle mesh Ewald (PME) method<sup>20</sup> was used to compute coulombic interactions and the grid spacings were about 1.0 Å. The PME interpolation order was cubic and the direct sum tolerance was  $10^{-6}$ . The water molecule geometry and covalent bonds between heavy and hydrogen atoms were fixed using SHAKE algorithm.<sup>21</sup> Full electrostatic evaluations were calculated every two time steps and multiple time step integration technique r-RESPA<sup>22</sup> was adopted. NPT ensembles were used where the pressure was set to 1.0 bar and the temperature was maintained to 300 K. The coupling methods for pressure and temperature were the same as that in other studies.<sup>2,23</sup> All simulations were done for 1 ns to equilibrate and then a production run of 1 ns were generated for analysis purpose. It should be noted that during the production run, several trajectory files generated from different time intervals were used. For hydrogen bonds kinetics analysis, trajectory files were generated every 5 time steps. For hydrogen bonding structure analysis, the average and error values are calculated from 400 files generated from a 200 ps production run with 0.5 ps interval each.

**Construction of Simulation Boxes:** An equilibrium water box was used to construct simulation boxes. According to the desired glucose concentrations, different numbers of glucose molecules were added into the water box at random positions with random velocities. Three simulation boxes with glucose mole fractions of 0.126, 0.151 and 0.223 were constructed.

**Hydrogen Bonds Definition.** In classical molecular dynamics simulations, several criteria for the definitions of hydrogen bonds by energy, geometry or topology have been proposed.<sup>24</sup> Geometry criterion has been extensively used because of its ability of exploring hydrogen bonds details like acceptor, donor and percentages. For a hydrogen bond X-H...A, the atom X is called donor atom and the atom A is called acceptor atom. Six parameters are involved in the hydrogen bond X-H...A: X...A distance, H...A hydrogen bond length, X-H covalent bond length, X-H...A hydrogen



**Figure 2.** (a) the intermolecular radial distribution functions  $g(r)$  for atom pairs OH2-OH, O1-OH, O2-OH, O3-OH, O4-OH and O6-OH as a function of distance  $r$  when glucose mole fraction is 0.126. (b) the intermolecular radial distribution functions  $g(r)$  for atom pairs H<sub>w</sub>-OH, HO1-OH, HO2-OH, HO3-OH, HO4-OH and HO6-OH as a function of distance  $r$  when glucose mole fraction is 0.126. H<sub>OH</sub> and O<sub>OH</sub> represents the hydrogen and oxygen atoms in six hydroxyl groups which are potentially donor and acceptor atoms in hydrogen bonds, respectively. The results for other glucose concentrations are similar and not shown here.

bond angle, H-X...A angle and X...A...H angle. Atoms H and A are defined as hydrogen bonded when the X...A distance or H...A distance is less than the threshold value and limitation to the angle X-H...A or H-X...A are often used concomitantly. In the present study, hydrogen bonds are defined on the basis of the H...A distance and H-X...A angle. The cutoff of H...O distance was usually selected from the position of the first minimum of the intermolecular oxygen-hydrogen radial distribution functions.

The radial distribution functions for atom pairs related with different hydroxyl groups have been calculated and the results have been shown in Figure 2. The positions of the first minima of the intermolecular oxygen-hydrogen radial distribution functions do not show important changes with hydroxyl groups and glucose concentrations. It's reasonable to use the same cutoff. We have taken 2.4 Å as the cutoff and results with 2.42 or 2.38 Å show the same trends as those reported here. The H-O...O angle cutoff for intermolecular and intramolecular hydrogen bonds were selected as 30° and 60°, respectively, as that in other studies.<sup>10</sup>

### Hydrogen Bonding Structure and Kinetics Analysis.

**Hydrogen Bonding Structure Analysis:** We define  $f_i^X$  ( $i = 0, 1, 2, 3, \dots$ ) as the percentage of oxygen atom  $X$  with  $i$  hydrogen bonds. In glucose aqueous solutions studied here, there are six oxygen atom types, namely OH2 in water molecules and O1, O2, O3, O4, O6 in glucose molecules. Similarly, we define  $f_i^Y$  ( $i = 0, 1, 2, 3, \dots$ ) as the percentage of hydrogen atom  $Y$  with  $i$  hydrogen bonds. There are six hydrogen atom types, namely H<sub>w</sub> in water molecules and HO1, HO2, HO3, HO4, HO6 in glucose molecules. It should be noted that the two hydrogen atoms in a water molecule have been regarded as one hydrogen atom type because of

the symmetrical geometry of water molecules used in the present study.  $n_X$  is referred to the mean number of hydrogen bonds per oxygen atom X and it can be calculated by the percentage  $f_i^X$ . Similarly,  $n_Y$  is referred to the mean number of hydrogen bonds per hydrogen atom Y which can be calculated by  $f_i^Y$ .

**Hydrogen Bonding Kinetics Analysis:** Hydrogen bonding kinetics has been analyzed by hydrogen bonding lifetimes. In the present study, hydrogen bonding lifetimes have been predicted by autocorrelation functions:<sup>25</sup>

$$\tau = \int_0^{\infty} C_{HB}(t) dt$$

where  $\tau$  is the hydrogen bonding lifetime and  $C_{HB}(t)$  is the autocorrelation function. The autocorrelation function can be expressed as:<sup>26-29</sup>

$$C_{HB}(t) = \frac{\langle p_{ij}(t) \cdot p_{ij}(0) \rangle}{\langle p_{ij}(0)^2 \rangle}$$

where  $p_{ij}(t)$  equals 0 or 1 depending on the hydrogen bond state of a given atoms pair  $i$  and  $j$ . If atoms  $i$  and  $j$  are hydrogen bonded at times 0 and  $t$  and the bond was never broken for periods longer than  $t^*$ ,  $p_{ij}(t) = 1$ . Otherwise,  $p_{ij}(t) = 0$ .

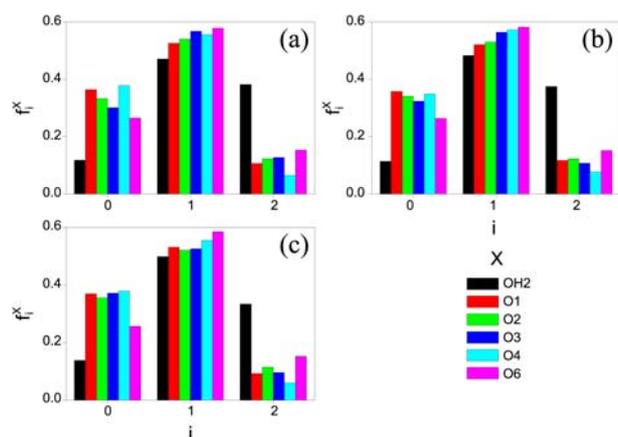
There are two limiting cases for  $t^*$ , so there are two autocorrelation functions namely the continuous autocorrelation function  $c_1(t)$  and the intermittent autocorrelation function  $c_2(t)$  corresponding to  $t^* = 0$  and  $t^* = \infty$ , respectively. To calculate the continuous autocorrelation function, the hydrogen bond states of atoms pairs must be analyzed at any time and the time step of molecular dynamics simulations has to be infinitely close to 0 which is impossible. In the present study, the hydrogen bond states were analyzed every 5 time steps and the continuous autocorrelation functions were calculated with  $t^* = 10$  fs. Corresponding to the continuous and intermittent autocorrelation functions, two hydrogen bonding lifetimes have been calculated namely the continuous hydrogen bonding lifetime  $\tau_1$  and the intermittent hydrogen bonding lifetime  $\tau_2$ , respectively.

## Results and Discussions

### Hydrogen Bonding Structure.

**O Atoms:** The percentages of different oxygen atoms have been calculated and the results have been illustrated in Figure 3. For OH2 atoms, in all concentrations studied, the percentage with one hydrogen bond is the largest and the percentage with no hydrogen bond is the smallest. However, for oxygen atoms in glucose molecules (O1, O2, O3, O4 and O6), the percentage with two hydrogen bonds is the smallest although the percentage with one hydrogen bond remains the largest.

When  $i = 1$ , the percentage of O6 is the largest and the percentage of OH2 is the smallest. The order for percentages of O1, O2, O3 and O4 changes with glucose concentrations. However, the percentage of O4 is larger than that of O1 and O2 and the percentage of O3 is larger than that of O2. When



**Figure 3.** Percentages of O atom types (OH2, O1, O2, O3, O4 and O6) with  $i$  hydrogen bonds when glucose mole fraction is 0.126 (a), 0.151 (b) and 0.223 (c).

$i = 2$ , the percentage of OH2 is the largest, the percentage of O6 is second largest and the percentage of O4 is the smallest. The order for percentages of O1, O2 and O3 changes with glucose mole fractions and no trend has been found. When  $i = 0$ , the percentage of OH2 is the smallest and the percentage of O6 is second smallest. The order for percentages of O1, O2, O3 and O4 changes with glucose concentrations. However, the percentage of O4 is larger than that of O2 and O3 and the percentage of O1 is larger than that of O2.

The mean numbers of hydrogen bonds per oxygen atom have been calculated and the results are summarized in Table 1. The mean number of hydrogen bonds per OH2 is larger than the mean numbers of hydrogen bonds per oxygen atom in glucose molecules, showing that the hydrogen bonding ability of OH2 is larger than that of oxygen atoms in glucose molecules. When glucose mole fraction is 0.126, the order for hydrogen bonding abilities of five oxygen atoms in glucose molecules is O6, O3, O2, O1 and O4 (from large to small). As glucose concentration increases to 0.151, the order changes to O6, O2, O3, O1 and O4. Further increasing of glucose concentration (0.223) doesn't change the order.

If we consider intermolecular hydrogen bonds, the mean number of hydrogen bonds per O6 is still the largest among the five glucose oxygen molecules. However, the order for hydrogen bonding abilities of other oxygen atoms is quite different from that when all hydrogen bonds have been considered. When glucose mole fraction is 0.126, the order for hydrogen bonding abilities from large to small is O3, O1, O2 and O4. The hydrogen bonding ability of O1 (O4) changes to be larger than the hydrogen bonding ability of O3 (O2) when glucose mole fraction increases to 0.151. If we further increase glucose mole fraction to 0.223, the hydrogen bonding ability of O4 becomes larger than that of O3.

For intramolecular hydrogen bonds, the hydrogen bonding abilities of O2 and O3 are the largest and second largest. The hydrogen bonding ability of O1, O4 and O6 decreases one by one when glucose mole fraction is 0.126. When glucose mole fraction increases to 0.151, the hydrogen bonding

**Table 1.** The mean numbers of hydrogen bonds per oxygen atom (OH2, O1, O2, O3, O4 and O6). “av” denotes the average value and “error” is the absolute deviation from the average value. The average and error values are calculated from 400 files generated from a 200 ps production run with 0.5 ps interval each

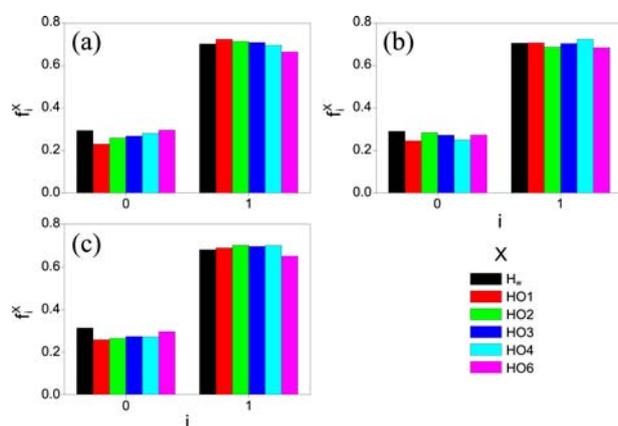
$x_s$	$n_{OH2}$		$n_{O1}$		$n_{O2}$		$n_{O3}$		$n_{O4}$		$n_{O6}$		
	av	error	av	error	av	error	av	error	av	error	av	error	
all	0.126	1.322	0.012	0.749	0.034	0.796	0.036	0.834	0.032	0.688	0.025	0.893	0.033
	0.151	1.317	0.011	0.768	0.028	0.793	0.029	0.790	0.026	0.729	0.026	0.892	0.027
	0.223	1.245	0.012	0.724	0.023	0.765	0.024	0.727	0.028	0.682	0.023	0.900	0.021
intra	0.126			0.034	0.007	0.131	0.017	0.087	0.015	0.031	0.009	0.018	0.006
	0.151			0.031	0.007	0.154	0.018	0.088	0.013	0.041	0.010	0.021	0.004
	0.223			0.023	0.005	0.155	0.015	0.096	0.014	0.041	0.009	0.032	0.005
inter	0.126			0.715	0.034	0.665	0.030	0.747	0.028	0.657	0.023	0.875	0.032
	0.151			0.737	0.027	0.639	0.024	0.702	0.022	0.688	0.028	0.871	0.027
	0.223			0.701	0.022	0.610	0.018	0.631	0.024	0.641	0.020	0.868	0.021

**Table 2.** The percentages of mean numbers of intermolecular ( $P_{inter}$ ) and intramolecular ( $P_{intra}$ ) hydrogen bonds per oxygen atom (O1, O2, O3, O4 and O6)

$x_s$	$n_{O1}$ (%)		$n_{O2}$ (%)		$n_{O3}$ (%)		$n_{O4}$ (%)		$n_{O6}$ (%)	
	$P_{inter}$	$P_{intra}$								
0.126	95.43	4.57	83.49	16.51	89.52	10.48	95.47	4.53	97.94	2.06
0.151	95.98	4.02	80.56	19.44	88.82	11.18	94.40	5.60	97.62	2.38
0.223	96.77	3.23	79.74	20.26	86.74	13.26	94.08	5.92	96.44	3.56

ability of O4 becomes larger than that of O1 and further increasing of glucose mole fraction to 0.223 makes the hydrogen bonding ability of O6 larger than that of O1.

The percentages of mean numbers of intermolecular and intramolecular hydrogen bonds per oxygen atom have been calculated and the results are summarized in Table 2. As can be seen, the percentage of intermolecular hydrogen bonds for O1 increases as glucose mole fraction increases. However, the percentage of intramolecular hydrogen bonds decreases as glucose concentration increases. For other oxygen atoms (O2, O3, O4 and O6), the percentages of intermolecular hydrogen bonds decrease while the percentages of intramolecular hydrogen bonds increase as glucose mole fraction increases.



**Figure 4.** Percentages of H atom types ( $H_w$ , HO1, HO2, HO3, HO4 and HO6) with  $i$  hydrogen bonds when glucose mole fraction is 0.126 (a), 0.151 (b) and 0.223 (c).

**H Atoms:** The percentages of different hydrogen atoms have been calculated and the results have been illustrated in Figure 4. For all hydrogen atoms, in all concentrations studied, the percentage with one hydrogen bond (over 60%) is larger than the percentage with no hydrogen bonds (below 40%). When  $i=1$ , the order for percentages of the six hydrogen atoms strongly depends on glucose concentrations. However, when  $i=0$ , the percentage of  $H_w$  is larger than that of hydrogen atoms in glucose molecules.

The mean numbers of hydrogen bonds per hydrogen atom have been calculated and the results are summarized in Table 3. The hydrogen bonding ability of  $H_w$  is smaller than the hydrogen bonding ability of hydrogen atoms in glucose molecules. Among the five hydrogen atoms in glucose molecules, the hydrogen bonding ability of HO1 is the largest. The order for hydrogen bonding abilities of other hydrogen atoms in glucose molecules is strongly dependent on glucose concentrations. If we consider intramolecular hydrogen bonds, the hydrogen bonding ability of HO1 is the largest and the hydrogen bonding ability of HO3 is the smallest. When intermolecular hydrogen bonds are considered, the order for hydrogen bonding abilities of hydrogen atoms in glucose molecules heavily correlates with glucose mole fractions.

The percentages of mean numbers of intermolecular and intramolecular hydrogen bonds per hydrogen atom have been calculated and the results have been summarized in Table 4. As can be seen, the percentages of intermolecular hydrogen bonds for HO2, HO3 and HO4 (over 90%) are larger than that for HO1 and HO6 (below 90%). Consequently, the percentages of intramolecular hydrogen bonds

**Table 3.** The mean numbers of hydrogen bonds per hydrogen atom ( $H_w$ , HO1, HO2, HO3, HO4 and HO6). “av” denotes the average value and “error” is the absolute deviation from the average value. The average and error values are calculated from 400 files generated from a 200 ps production run with 0.5 ps interval each

$x_s$	$n_{Hw}$		$n_{HO1}$		$n_{HO2}$		$n_{HO3}$		$n_{HO4}$		$n_{HO6}$		
	av	error	av	error	av	error	av	error	av	error	av	error	
all	0.126	0.709	0.009	0.818	0.025	0.771	0.023	0.758	0.024	0.745	0.024	0.747	0.029
	0.151	0.712	0.009	0.805	0.023	0.747	0.022	0.754	0.022	0.777	0.024	0.773	0.024
	0.223	0.688	0.011	0.795	0.021	0.770	0.021	0.757	0.022	0.756	0.018	0.757	0.022
intra	0.126			0.108	0.015	0.057	0.012	0.044	0.011	0.067	0.013	0.106	0.017
	0.151			0.139	0.017	0.058	0.012	0.050	0.013	0.059	0.011	0.102	0.017
	0.223			0.133	0.014	0.058	0.013	0.058	0.011	0.072	0.010	0.115	0.015
inter	0.126			0.710	0.021	0.714	0.021	0.714	0.023	0.678	0.021	0.641	0.023
	0.151			0.666	0.022	0.689	0.019	0.704	0.024	0.718	0.021	0.671	0.019
	0.223			0.662	0.017	0.712	0.020	0.699	0.020	0.684	0.017	0.642	0.019

**Table 4.** The percentages of mean numbers of intermolecular ( $P_{inter}$ ) and intramolecular ( $P_{intra}$ ) hydrogen bonds per hydrogen atom (HO1, HO2, HO3, HO4 and HO6)

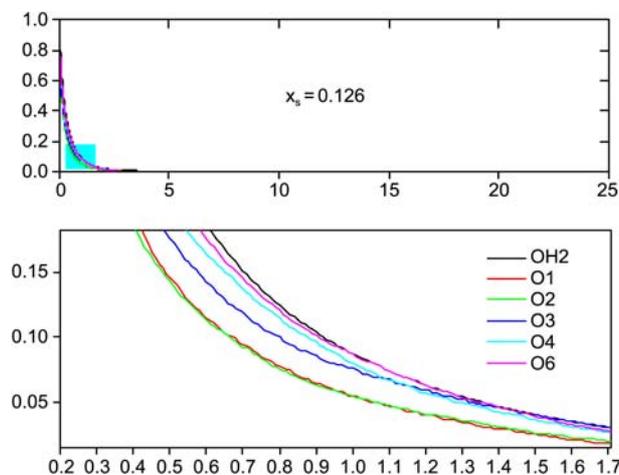
$x_s$	$n_{HO1}$ (%)		$n_{HO2}$ (%)		$n_{HO3}$ (%)		$n_{HO4}$ (%)		$n_{HO6}$ (%)	
	$P_{inter}$	$P_{intra}$								
0.126	86.80	13.20	92.61	7.39	94.20	5.80	91.01	8.99	85.81	14.19
0.151	82.73	17.27	92.24	7.76	93.37	6.63	92.41	7.59	86.80	13.20
0.223	83.27	16.73	92.47	7.53	92.34	7.66	90.48	9.52	84.81	15.19

for HO1 and HO6 are larger than that for HO2, HO3 and HO4.

### Hydrogen Bonding Kinetics.

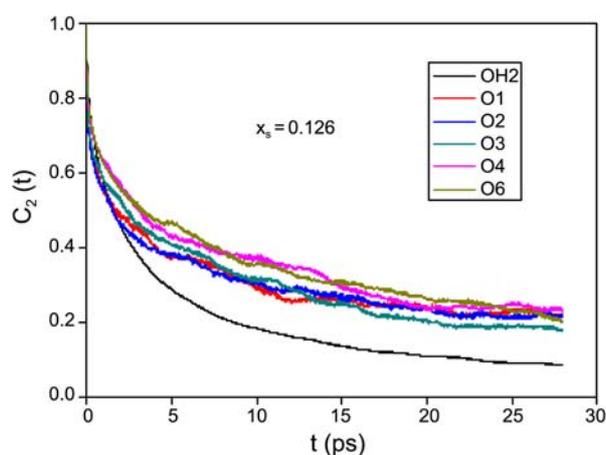
**O Atoms:** The continuous and intermittent autocorrelation functions for hydrogen bonds associated with OH2, O1, O2, O3, O4 and O6 have been calculated and the results have been illustrated in Figures 5 and 6, respectively. Only the results when glucose mole fraction is 0.126 have been shown in the text and the results for other glucose concentrations have been summarized in the supplementary file (Figures S1-S4). It can be seen that, the autocorrelation function  $c_2(t)$  strongly depends on oxygen atom types while  $c_1(t)$  shows a little dependence on oxygen atom types. The continuous autocorrelation functions decrease sharply as time goes on. When glucose mole fraction is 0.126, after 0.19, 0.12, 0.10, 0.12, 0.15 and 0.16 ps, the continuous autocorrelation functions for OH2, O1, O2, O3, O4 and O6 have dropped from 1 to 0.5. The intermittent autocorrelation functions also decrease with time but the decreasing is quite flat. For intermittent autocorrelation functions, the time intervals required to decrease from 1 to 0.5 are 1.58, 1.64, 1.60, 2.37, 3.07 and 3.41 ps, respectively. The results are similar for other glucose concentrations studied.

The continuous hydrogen bonding lifetime  $\tau_1$  and the intermittent hydrogen bonding lifetime  $\tau_2$  for hydrogen bonds related with OH2, O1, O2, O3, O4 and O6 have been predicted using the continuous and intermittent autocorrelation functions, respectively and the results have been listed in Table 5. The continuous hydrogen bonding lifetime lasts until the first rupture of hydrogen bonds and it represents the hydrogen bonding strength. The continuous hydrogen bond-



**Figure 5.** The top part: the continuous autocorrelation functions  $c_1(t)$  (vertical) for hydrogen bonds associated with OH2, O1, O2, O3, O4 and O6 as a function of time (horizontal, in units of ps) when glucose mole fraction is 0.126. The bottom part: the top part has been enlarged locally.

ing lifetimes show a tendency of increase as glucose mole fraction increases, although there are several exceptions for O1 and O3 when glucose mole fraction is 0.151. The increasing of continuous hydrogen bonding lifetimes of oxygen atoms in glucose molecules seems faster than that of OH2. When glucose mole fraction is below 0.151, the continuous hydrogen bonding lifetime of OH2 is larger than that of oxygen atoms in glucose molecules. However, when glucose mole fraction increases to 0.223, the continuous hydrogen bonding lifetime of O6 becomes larger than that of



**Figure 6.** The intermittent autocorrelation functions  $c_2(t)$  for hydrogen bonds associated with OH2, O1, O2, O3, O4 and O6 as a function of time when glucose mole fraction is 0.126.

**Table 5.** The continuous and intermittent hydrogen bonding lifetimes  $\tau_1$  and  $\tau_2$  for hydrogen bonds related with OH2, O1, O2, O3, O4 and O6. The errors are 0.02 ps

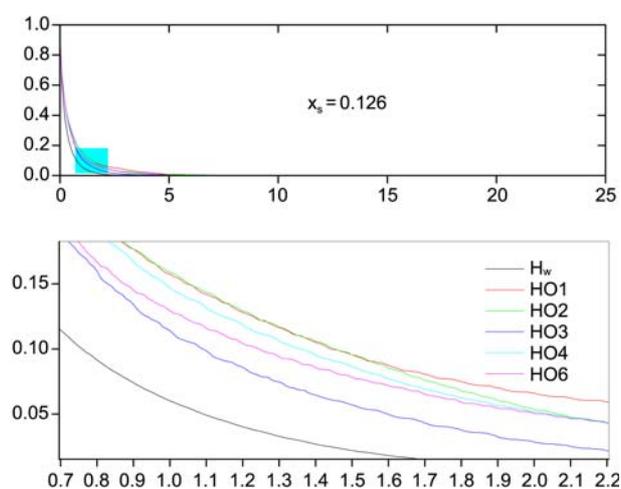
	$x_s$	OH2	O1	O2	O3	O4	O6
$\tau_1$ /ps	0.126	0.38	0.28	0.26	0.31	0.33	0.35
	0.151	0.43	0.24	0.27	0.25	0.34	0.35
	0.223	0.45	0.44	0.33	0.31	0.39	0.53
$\tau_2$ /ps	0.126	5.55	8.51	8.46	8.37	9.66	9.77
	0.151	6.55	8.69	9.80	8.06	10.11	10.34
	0.223	8.20	12.36	12.63	12.31	13.55	13.84

OH2. Among the five oxygen atoms in glucose molecules, the continuous hydrogen bonding lifetime of O6 is the largest. The continuous hydrogen bonding lifetime of O4 is larger than that of O2 and O3.

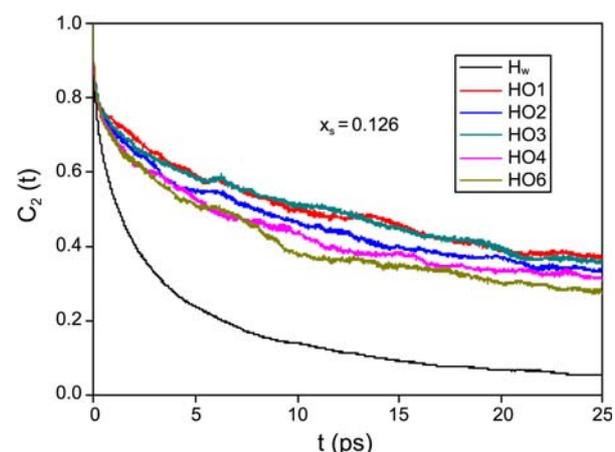
The intermittent hydrogen bonding lifetime lasts until one hydrogen bond forms again ignoring the intermediate status. The intermittent hydrogen bonding lifetime increases as glucose mole fraction increases. There is an exception for O3 when glucose mole fraction is 0.151. Unlike the continuous hydrogen bonding lifetime, the intermittent hydrogen bonding lifetime of OH2 is smaller than that of oxygen atoms in glucose molecules. In the five oxygen atoms in glucose molecules, the intermittent hydrogen bonding lifetimes of O6 and O4 are the largest and second largest, respectively. The intermittent hydrogen bonding lifetime of O1 is larger than that of O3.

The intermittent hydrogen bonding lifetimes are larger than the continuous hydrogen bonding lifetimes. For OH2, the ratio of  $\tau_2$  to  $\tau_1$  is between 14.6 and 18.2. This is consistent with other studies where a factor of 5-20 at ambient conditions and 3-7 at supercritical region for water has been reported.<sup>30</sup> The ratios for O1, O2, O3, O4 and O6 are 28.1-36.2, 32.5-38.3, 27-39.7, 29.3-34.7 and 26.1-29.5, respectively.

**H Atoms:** The continuous and intermittent autocorrelation functions for hydrogen bonds associated with  $H_w$ , HO1,



**Figure 7.** The top part: the continuous autocorrelation functions  $c_1(t)$  (vertical) for hydrogen bonds associated with  $H_w$ , HO1, HO2, HO3, HO4 and HO6 as a function of time (horizontal, in units of ps) when glucose mole fraction is 0.126. The bottom part: the top part has been enlarged locally.



**Figure 8.** The intermittent autocorrelation functions  $c_2(t)$  for hydrogen bonds associated with  $H_w$ , HO1, HO2, HO3, HO4 and HO6 as a function of time when glucose mole fraction is 0.126.

HO2, HO3, HO4 and HO6 have been calculated and the results have been illustrated in Figures 7 and 8, respectively. Only the results when glucose mole fraction is 0.126 have been shown in the text and the results for other glucose concentrations have been summarized in the supplementary file (Figures S5-S8). Similar to the autocorrelation functions for hydrogen bonds related with oxygen atoms, the autocorrelation functions for hydrogen bonds related with hydrogen atoms decrease as time goes on and the decreasing of  $c_1(t)$  is faster than that of  $c_2(t)$ .

The continuous hydrogen bonding lifetime  $\tau_1$  and the intermittent hydrogen bonding lifetime  $\tau_2$  for hydrogen bonds related with  $H_w$ , HO1, HO2, HO3, HO4 and HO6 have been predicted using the continuous and intermittent autocorrelation functions, respectively and the results have been listed in Table 6.

The continuous hydrogen bonding lifetimes for  $H_w$  and

**Table 6.** The continuous and intermittent hydrogen bonding lifetimes  $\tau_1$  and  $\tau_2$  for hydrogen bonds related with  $H_w$ , HO1, HO2, HO3, HO4 and HO6. The errors are 0.02 ps

	$x_s$	$H_w$	HO1	HO2	HO3	HO4	HO6
$\tau_1$ /ps	0.126	0.30	0.57	0.53	0.44	0.51	0.50
	0.151	0.30	0.66	0.50	0.59	0.51	0.45
	0.223	0.31	0.59	0.50	0.47	0.73	0.58
$\tau_2$ /ps	0.126	4.33	13.60	12.51	13.46	11.78	11.08
	0.151	5.00	13.74	12.26	12.42	13.38	13.05
	0.223	6.17	16.81	16.07	15.20	16.03	15.47

HO4 increase as glucose concentration increases. Unlike hydrogen bonds related with oxygen atoms, the continuous hydrogen bonding lifetime for  $H_w$  is smaller than that for hydrogen bonds related with hydrogen atoms in glucose molecules. The hydrogen bonding strength for hydrogen atoms in glucose molecules change fast with glucose concentration. However, the continuous hydrogen bonding lifetime for HO1 is larger than that for HO2 and HO3 and the continuous hydrogen bonding lifetime for HO4 is larger than that for HO6.

The intermittent hydrogen bonding lifetime increases as glucose mole fraction increases. There are exceptions for HO2 and HO3 when glucose mole fraction is 0.151. The same as the continuous hydrogen bonding lifetime, the intermittent hydrogen bonding lifetime of  $H_w$  is smaller than that of hydrogen atoms in glucose molecules. In the five hydrogen atoms in glucose molecules, the intermittent hydrogen bonding lifetimes of HO1 is the largest. The ratio between  $\tau_2$  and  $\tau_1$  has also been calculated. For  $H_w$ , the ratio is between 14.4 and 19.9. For HO1, HO2, HO3, HO4 and HO6, the ratios are 20.8-28.5, 23.6-32.1, 21.1-32.3, 21.9-26.2 and 22.2-29, respectively.

#### Discussions.

**Acceptor and Donor:** For a hydroxyl group, it acts as an acceptor when its hydrogen atom is involved with hydrogen bonds and it acts as a donor when its oxygen atom is involved with hydrogen bonds. The hydrogen bonding abilities for different oxygen and hydrogen types have been analyzed in sections 3.1. The results can be concluded as follows:

1) The hydrogen bonding ability of OH2 is larger than that of O1, O2, O3, O4 and O6. Among the five O in glucose molecules, the hydrogen bonding ability of O6 is the largest and the hydrogen bonding ability of O4 is the smallest. O6 has the largest ability to form intermolecular hydrogen bonds while O2 and O3 have the largest and second largest abilities to form intramolecular hydrogen bonds.

2) The hydrogen bonding ability of  $H_w$  is smaller than that of HO1, HO2, HO3, HO4 and HO6. The hydrogen bonding ability of HO1 is the largest among the five H in glucose molecules. HO1 and HO3 have the largest and the smallest abilities to form intermolecular hydrogen bonds, respectively.

According to the hydrogen bonding abilities analysis of different O and H atoms: OH2- $H_w$  is the best acceptor and

the poorest donor; among the five hydroxyl groups in glucose, O6-HO6 is the best acceptor and poorer donor, O1-HO1 is the best donor and O4-HO4 is the poorest acceptor. A part of the results (O6-HO6 is the best acceptor and O1-HO1 is the best donor) are in good agreement with the results found by other researchers.<sup>11</sup> The donor and acceptor abilities of hydroxyl groups change with concentrations. The glucose concentration in Ref. [11] is quite small and different from concentrations in the present study. It should be noted that the hydrogen bonding abilities of O(H) in glucose aqueous solutions are smaller than that in glycerol aqueous solutions.<sup>2</sup> In fact, there are total five hydroxyl groups in glucose; however, there are only three hydroxyl groups in glycerol. The competition between hydroxyl groups decreases the hydrogen bonding abilities of O(H) in glucose aqueous solutions. The reasons for different hydrogen bonding abilities for oxygen and hydrogen types are complicated and further investigation is required in future.

**Acceptor/Donor Efficiency:** The acceptor/donor efficiency can be estimated by the ratio of the number of hydrogen bonds as acceptors to the number of hydrogen bonds as donors.

**Molecules** – For water molecules, the ratios at all concentrations studied are all larger than one showing that water molecules are more efficient acceptors than donors. For glucose molecules, the ratios are larger than one (1.03) when glucose mole fraction is 0.126 and 0.151, however, the ratio becomes to smaller than one (0.99) when glucose mole fraction is 0.223. The results are consistent with that in literature. It has been showed that glucose molecules are more efficient donors than acceptors during a study of glucose aqueous solutions with concentrations of 1, 3 and 5 m.<sup>12</sup> The glucose concentrations in the present study are 0.80, 0.99 and 1.59 m, respectively. It may be concluded that the acceptor/donor efficiency of glucose molecules decreases as glucose concentration increases. At low glucose concentrations, glucose molecules are more efficient acceptors than donors and at high glucose concentrations, glucose molecules are more efficient donors than acceptors. Based on the results in the present study, the transition glucose mole fraction may occur around 0.151. However, the assumption must be verified by investigating glucose aqueous solutions with wide concentration range (such as mole fraction from 0 to 1) and this will be done in future studies. If only intramolecular hydrogen bonds are considered, the acceptor/donor efficiencies of glucose molecules are all smaller than one showing that glucose molecules are more efficient intramolecular hydrogen bonding donors than acceptors. If only intermolecular hydrogen bonds are considered, the acceptor/donor efficiencies of glucose molecules are all larger than one (1.058, 1.055 and 1.015 for the three concentrations studied). However, the ratios are quite close to one implying that for higher glucose mole fractions (larger than 0.223), glucose molecules may be more efficient intermolecular hydrogen bonding donors than acceptors.

**Hydroxyl Groups** – The acceptor/donor efficiencies of hydroxyl groups have also been analyzed. For OH2- $H_w$

(only one hydrogen atom is included here) and O6-HO6, the ratios are all larger than 1 showing that these two hydroxyl groups are more efficient hydrogen bonding acceptors than donors. The ratios of O1-HO1 and O4-HO4 are smaller than one implying that these two hydroxyl groups are more efficient hydrogen donors than acceptors. The ratios of O2-HO2 and O3-HO3 are larger than one when glucose mole fraction is below 0.151. However, when glucose mole fraction increases to 0.223, the ratios become smaller than one. The behavior of O2-HO2 and O3-HO3 is the same as that of overall glucose molecules.

If only intramolecular hydrogen bonds are considered, the ratios of O2-HO2 and O3-HO3 are larger than 2 and 1.5, respectively. It can be concluded that O2-HO2 and O3-HO3 are more efficient intramolecular hydrogen bonding acceptors than donors. The ratios of O1-HO1, O4-HO4 and O6-HO6 are all smaller than one (the ratio of O6-HO6 is as low as 0.2) showing that these hydroxyl groups are more efficient intramolecular hydrogen bonding donors than acceptors.

If only intermolecular hydrogen bonds are considered, the ratios of O1-HO1 and O6-HO6 are larger than one while the ratios of O2-HO2 and O4-HO4 are smaller than one. The ratio of O3-HO3 is larger than one when glucose mole fraction is 0.126. The ratio becomes smaller than one when glucose concentration further increases. It can be concluded that hydroxyl groups O1-HO1 and O6-HO6 are more efficient intermolecular hydrogen bonding acceptors than donors while hydroxyl groups O2-HO2 and O4-HO4 are more efficient intermolecular hydrogen bonding donors than acceptors. At first, the hydroxyl group O3-HO3 seems to be more efficient intermolecular hydrogen bonding acceptors than donors and as glucose concentration increases O3-HO3 becomes more efficient intermolecular hydrogen bonding donors than acceptors.

**Hydrogen Bonding Abilities of Hydroxyl Groups:** For a hydroxyl group, its hydrogen bonding ability can be evaluated by summing the mean number of hydrogen bonds of its O and H atoms. The hydrogen bonding ability of OH2-H<sub>w</sub> (only one hydrogen atom is included here) is about 2.0 and the hydrogen bonding ability of hydroxyl groups in glucose falls between 1.4 and 1.7. Among the five hydroxyl groups in glucose, the hydrogen bonding ability of O6-HO6 is the largest and the hydrogen bonding ability of O4-HO4 is the smallest. If only intermolecular hydrogen bonds are considered, the hydrogen bonding ability of O6-HO6 is the largest while the hydroxyl group which hydrogen bonding ability is the smallest changes with glucose concentrations. If only intramolecular hydrogen bonds are considered, the order for hydrogen bonding abilities from large to small is O2-HO2, O1-HO1, O3-HO3, O6-HO6 and O4-HO4.

### Conclusions and Outlooks

Molecular dynamics simulations have been performed to investigate hydrogen bonding characteristics of hydroxyl groups in glucose aqueous solutions with different concentrations.

The mean numbers of hydrogen bonds (all, intermolecular, intramolecular) per atom for different O (OH2 in water, O1, O2, O3, O4 and O6 in glucose) and H (H<sub>w</sub> in water, HO1, HO2, HO3, HO4 and HO6 in glucose) have been calculated to assess the hydrogen bonding abilities of these different atom types. The hydrogen bonding lifetimes for hydrogen bonds associated with different O(H) types have also been predicted to investigate the hydrogen bonding strength. It has been found that: (1) the hydrogen bonding ability and strength of OH2 are larger than that of O in glucose while the hydrogen bonding ability and strength of H<sub>w</sub> are smaller than that of H in glucose; (2) among the five O in glucose, the hydrogen bonding ability and strength of O6 are the largest, O4 has the smallest hydrogen bonding ability, O6 has the largest ability to form intermolecular hydrogen bonds while O2 and O3 have the largest and second largest abilities to form intramolecular hydrogen bonds; (3) among the five H in glucose, the hydrogen bonding ability of HO1 is the largest, HO1 and HO3 have the largest and the smallest abilities to form intermolecular hydrogen bonds. The reasons for different hydrogen bonding abilities for oxygen and hydrogen types are complicated and further investigation is required in future.

The acceptor/donor efficiencies have been predicted by the ratio of the number of hydrogen bonds as acceptors to the number of hydrogen bonds as donors. The hydrogen bonding abilities of hydroxyl groups have also been calculated. In the present study, the hydrogen bonding abilities and strength as well as acceptor/donor efficiencies have been found to be concentration dependent. They may be temperature dependent. The exact mechanisms for the differences of hydroxyl groups are open to questions. The glucose conformations and hydrogen bonding characteristics such as electron transfer may be helpful to answer these questions and should be considered in further research.

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### References

1. Mazur, P. *Am. J. Physiol.* **1984**, *247*, C125.
2. Chen, C.; Li, W. Z.; Song, Y. C.; Yang, J. *J. Mol. Liq.* **2009**, *146*, 23.
3. Sugimachi, K.; Roach, K. L.; Rhoads, D. B.; Tompkins, R. G.; Toner, M. *Tissue Eng.* **2006**, *12*, 579.
4. Holmstrup, M.; Overgaard, J.; Bindesbol, A. M.; Pertoldi, C.; Bayley, M. *Soil Biology & Biochemistry* **2007**, *39*, 2640.
5. Dinsmore, S. C.; Swanson, D. L. *Canadian Journal of Zoology- Revue Canadienne De Zoologie* **2008**, *86*, 1095.
6. Martinez, V. Y.; Nieto, A. B.; Castro, M. A.; Salvatori, D.; Alzamora, S. M. *J. Food Eng.* **2007**, *83*, 394.
7. Lee, S. L.; Debenedetti, P. G.; Errington, J. R. *J. Chem. Phys.*

- 2005; p 122 .
- Paolantoni, M.; Sassi, P.; Morresi, A.; Santini, S. *J. Chem. Phys.* **2007**, *127*, 024504.
  - Te, J. A.; Tan, M. L.; Ichiye, T. *Chem. Phys. Lett.* **2010**, *491*, 218.
  - Chen, C.; Li, W.-Z.; Song, Y.-C.; Weng, L.-D. *Acta Physico-chimica Sinica* **2011**, *27*, 1372.
  - Suzuki, T. *PCCP* **2008**, *10*, 96.
  - Mason, P. E.; Neilson, G. W.; Enderby, J. E.; Saboungi, M. L.; Brady, J. W. *J. Phys. Chem. B* **2005**, *109*, 13104.
  - Max, J. J.; Chapados, C. *J. Phys. Chem. A* **2007**, *111*, 2679.
  - Paolantoni, M.; Comez, L.; Fioretto, D.; Gallina, M. E.; Morresi, A.; Sassi, P.; Scarponi, F. *Journal of Raman Spectroscopy* **2008**, *39*, 238.
  - Aroulmoji, V.; Mathlouthi, M.; Feruglio, L.; Murano, E.; Grassi, M. *Food Chem.* 2011.
  - Venable, R. M.; Hatcher, E.; Guvench, O.; Alexander, J.; MacKerell, D.; Pastor, R. W. *J. Phys. Chem. B* **2010**, *114*, 12501.
  - Chen, C.; Li, W. Z.; Song, Y. C.; Weng, L. D.; Zhang, N. *Computational and Theoretical Chemistry* **2012**, *984*, 85.
  - Phillips, J. C.; Braun, R.; Wang, W.; Gumbart, J.; Tajkhorshid, E.; Villa, E.; Chipot, C.; Skeel, R. D.; Kale, L.; Schulten, K. *J. Comput. Chem.* **2005**, *26*, 1781.
  - Guvench, O.; Greene, S. N.; Kamath, G.; Brady, J. W.; Venable, R. M.; Pastor, R. W.; Alexander, J.; MacKerell, D. *J. Comput. Chem.* **2008**, *29*, 2543.
  - Darden, T.; York, D.; Pedersen, L. *J. Chem. Phys.* **1993**, *98*, 10089.
  - Ryckaert, J. P. *Mol. Phys.* **1985**, *55*, 549.
  - Tuckerman, M.; Berne, B. J.; Martyna, G. J. *The Journal of Chemical Physics* **1992**, *97*, 1990.
  - Weng, L. D.; Chen, C.; Zuo, J. G.; Li, W. Z. *J. Phys. Chem. A* **2011**, *115*, 4729.
  - Henchman, R. H.; Irudayam, S. J. *The Journal of Physical Chemistry B* **2010**, *114*, 16792.
  - Skarmoutsos, L.; Guardia, E.; Samios, J. *The Journal of Chemical Physics* **2010**, *133*, 014504.
  - Elola, M. D.; Ladanyi, B. M. *The Journal of Chemical Physics* **2006**, *125*, 184506.
  - Lee, H.-S.; Tuckerman, M. E. *The Journal of Chemical Physics* **2007**, *126*, 164501.
  - Guardia, E.; Marti, J.; Padro, J. A.; Saiz, L.; Komolkin, A. V. *Journal of Molecular Liquids* **2002**, *96-97*, 3.
  - Root, L. J.; Berne, B. J. *J. Chem. Phys.* **1997**, *107*, 4350.
  - Petrenko, V. E.; Antipova, M. L. *Structural Chemistry* **2011**, *22*, 471.
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