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Molecular dynamics simulations of α-D-Man*p*-(1→3)-β-D-Glc*p*-OMe in methanol and in dimethyl sulfoxide solutions

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Molecular dynamics simulations have been performed of the disaccharide α-D-Manp-(1→3)-β-D-Glcp-OMe in two different solvents, namely in methanol and in dimethyl sulfoxide. The conformation of the disaccharide is similar to that previously determined in water. The three-dimensional structure around the solute was investigated by geometric hydrogen bonding criteria, radial distribution functions, coordination number analysis, residence times for hydrogen bonds, and spatial distribution functions. Differences and similarities between methanol and the aprotic dimethyl sulfoxide as solvent are analyzed. © 2001 by Elsevier Science Inc.

Keywords: disaccharide, hydrogen bonding, radial distribution function, spatial distribution function

INTRODUCTION

Carbohydrates, being one subclass of biomolecules, are intimately linked to lipids and proteins, usually as glycoconjugates, e.g., glycolipids in membranes or as glycoproteins. The molecules in these complex biomolecular systems interact with each other, leading to the regulation of for example, the duration of a glycoprotein in serum, the binding of bacterial toxins to a cell surface, or the initial adhesion of cells to one another.[1,2] In these studies experimental techniques, such as X-ray crystallography[3] and NMR spectroscopy,[4] are important for investigating structure. Molecular modeling of carbohydrates is usually performed as parts of the above studies. However, techniques based on Monte Carlo (MC), molecular dynamics (MD), or other types of simulations can on their own advance the knowledge of these molecular systems. Together with computer graphics visualization, these simulation techniques offer a powerful tool for investigating the three-dimensional structure and dynamics in complex molecular systems, such as carbohydrates. MD simulations of oligosaccharides in solution have revealed that structuring around the solute in water is highly anisotropic.[5,6] Furthermore, the importance of solvent on the conformational equilibrium has been highlighted in these studies. In binary solvent mixtures, different components have shown preferential solvation for different saccharide hydroxyl groups. This was investigated using the well-known radial distribution functions (RDF) but also the newer spatial distribution functions (SDF)[7,8] in three dimensions, where the angular dependence of the interaction is included. The disaccharide α-D-Man*p*-(1→3)-β-D-Glc*p*-OMe, a model similar in structure to disaccharide elements found in glycoproteins and polysaccharides, (for which we previously reported carbon-13 relaxation studies in the binary mixture water:dimethyl sulfoxide (7:3)[9] and MD simulations in a 3:1 mixture,[10]) is here investigated by MD simulations in the two solvents methanol and dimethyl sulfoxide (DMSO), in particular with respect to solvent ordering around the solute disaccharide.

MODELS AND SIMULATION DETAILS

The force field model used for the disaccharide was the GLYCAM_93 parameter set,[11] based on the AMBER force field.

Color Plates for this article are on pages 396–397.

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Partial charges for use with the GLYCAM parameter set were calculated at the RHF/6-31G* level with the Gaussian 94 package. The complete set is described in our previous article.[6] For the present study, we chose the three-site model of van Leeuwen and Smit for methanol[12] and the P2 model of Luzar and Chandler for DMSO.[13] The heat of evaporation and the diffusion coefficient for liquid DMSO, obtained with the P2 model, is in good agreement with experimental data.[14]

In the initial part of the simulation, the disaccharide molecule was placed in a cavity, surrounded by 256 solvent molecules arranged as a face-centered cubic lattice with a density of 0.6 g/cm^3. During the first 10 ps of trajectories the volume of the cubic simulation box was gradually compressed to the experimental density of the solvent. This was followed by a 40-ps NVT simulation, keeping the solute molecule rigid, while a preliminary equilibrium in the solvent was established. Thereafter, the simulations were carried out in the NPT ensemble at P = 1.0 atm and T = 288 K, both in the methanol solution and in DMSO solution. The systems were simulated for 1.27 and 1.15 ns with $\rho = 0.77$ in methanol and $\rho = 1.15$ g/cm^3 in DMSO. Statistics were collected from 190 ps to the end of each trajectory. Methanol and DMSO were kept rigid using the SHAKE algorithm.[15] The temperature and pressure were maintained by the Nóse-Hoover thermostat and barostat, respectively.[16,17] For the flexible disaccharide, the multiple time step method by Tuckerman et al. was used in the simulations.[18] Fast fluctuating forces from the closest nonbonded interactions, stretching of bonds, and covalent angles were calculated in intervals of 0.2 fs, while a longer time step of 1.0 fs was used for other interactions. All the simulations were carried out on a dual 300-MHz PentiumII, running Linux, using the general purpose simulation software M.Dyna*Mix*.[19]

The atoms of the mannosyl group are designated with an *m* and those in the glucosyl residue by a *g*. The torsion angles across the glycosidic linkage are defined for ϕ as H1*m*-C1*m*-O3*g*-C3*g* and for ψ as H3*g*-C3*g*-O3*g*-C1*m*. For investigation of the three-dimensional solvent structure around the disaccharide, the molecular reference coordinate system was fixed by two vectors, namely O3*g*-C1*g* and O3*g*-C5*m*.

RESULTS AND DISCUSSION

Disaccharide Conformation

The conformational preference of a disaccharide is usually described by the torsion angles at the glycosidic linkage, namely ϕ and ψ (Figure 1). During the ~1 ns simulations of α-D-Man*p*-(1→3)-β-D-Glc*p*-OMe in the solvents methanol and in DMSO, only one conformational state is populated. The average torsion angles are quite similar in the two solvents with ϕ/ψ averages of −31°/58° in methanol and −30°/61° in DMSO, respectively. These average torsion angles and flexibility are very close to those obtained in water using the same GLYCAM force field that was used in our previous simulations.[6] We previously exploited the combination of GLYCAM for the sugar together with different solvent models for the mixture, such as the P2 model for DMSO combined, with SPC water. However, the mixed solvent P2/SPC showed the ϕ torsion angle of the disaccharide to be staggered and positive, which is not the major conformational state, while another combination of models, OPLS/TIP3P, for the mixed solvent leads to a conformational state that is expected to be the major

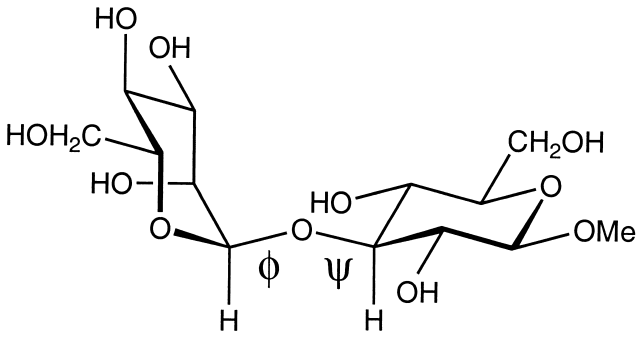

Figure 1. Schematic of α-D-Manp-(1→3)-β-D-Glcp-OMe with the glycosidic torsion angles.

or at least a significantly occupied state. For possible future comparisons to experiment, the *trans*-glycosidic distance *r* for H1*m*-H3*g* was calculated: in methanol $\langle r \rangle = 2.34$ Å and in DMSO $\langle r \rangle = 2.38$ Å, both distances being quite similar to those calculated in other solvents. Having identified that the disaccharide in these simulations shows one conformational state, we continue to analyze the interaction between solute and solvent.

Hydrogen Bonding

Characteristics of the intermolecular solute–solvent hydrogen bonds are compiled in Table 1. In general, the donation of hydroxyl hydrogens of the solute is similar in the two solvents for each hydroxyl group. Decreased donor ability is observed in particular for HO6*m*. In this region of the solute molecule, intramolecular hydrogen bonds have been observed in the previous simulations. In the present simulations hydrogen bonding occurs between HO6*m* and O2*g* 77% of the time in methanol and 56% in DMSO. A reduced hydrogen bond formation to solvent caused by an intramolecular hydrogen bond was also reported by Berger et al. for sucrose in DMSO.[20] As in water, when dissolved in methanol the solute can also accept

Table 1. Trajectory-averaged number of hydrogen bonds between solvent and selected hydroxyl groups in the disaccharide. Geometric criteria: donor-hydrogen· · ·acceptor angle ⩾ 120° and distance ⩽ 3.4 Å

Oxygen atom	Methanol donated	Methanol accepted	DSMO donated
O5*m*	–	0.03	–
O2*m*	0.47	0.97	0.59
O3*m*	0.68	0.92	0.52
O4*m*	0.59	1.11	0.27
O6*m*	0.05	1.02	0.05
O5*g*	–	0.16	–
O2*g*	0.46	0.21	0.80
O3*g*	–	0.04	–
O4*g*	0.77	1.03	0.92
O6*g*	0.72	0.89	0.85
O1*g*	–	0.33	–

hydrogen bonds from the solvent. For nonhydroxyl oxygens this occurs to a lower extent, but for the hydroxyl groups roughly one hydrogen bond per group is accepted in the methanol solvent. Again, an exception occurs, namely for O2*g* where the donation from solvent is low. This is not surprising since an intramolecular hydrogen bond is formed from HO6*m* as described above.

Radial Distribution Functions

Solvent structure around the solute has traditionally been studied using radial distribution functions (RDF) of atom density given by:

$$g_{AB}(r) = \frac{\rho_B(r|r_A = 0)}{\rho_B} \quad (1)$$

where the first index A refers to a solute atom at the origin, and the second index B refers to a solvent atom B, r is the distance between the atoms, and ρ_B is the bulk density of B in the solvent. Three types of RDFs were calculated: $g_{OO}(r)$, $g_{OH}(r)$, and $g_{HO}(r)$. Coordination numbers were obtained by integrating $g_{AB}(r)$ out to the first minimum. These were 3.7 Å $g_{OO}(r)$ in both solvents, 2.5 Å for $g_{OH}(r)$ in methanol, and 2.8 Å and 3.2 Å for $g_{HO}(r)$ in methanol and DMSO, respectively (Table 2).

The $g_{OO}(r)$ radial distributions in methanol show for most RDFs the first maximum around 2.7 Å, whereas for DMSO it is found around 2.9 Å. Only in a few cases, nonexistent or low intensity maxima occur at these distances. In methanol this is observed for O2*g* and in DMSO for O4*m* and O6*m* giving low coordination numbers. A conspicuous difference between the two solvents is observed for O6*m* (Figure 2a). For $g_{OH}(r)$ distributions in methanol the first maximum occurs around 1.7 Å with coordination numbers close to 2 indicating that two solvent molecules act as donors of hydrogen bonds to the solute. The exception is found for O2*g*, which gives a low coordination number of 0.6, which may be compared with a coordination number of 1.6 for O2*m* (Figure 2b). When $g_{HO}(r)$ in methanol is analyzed a well-defined second maximum around 3.3 Å is common in methanol (Figure 2c). In DMSO, no well-developed second maximum is observed (Figure 2d). Similar in the two solvents is the presence of a first maximum at ~2 Å for $g_{HO}(r)$ of HO6*g*, which is absent for HO6*m*. These results reveal different structures in the first coordination shell as a consequence of the different properties of the two solvents. The second maximum of $g_{HO}(r)$ in methanol is due to the proximity of another solvent molecule acting as a donor. In DMSO, the coordination numbers n_{HO} are in most cases twice as large, presumably because the oxygen atoms of DMSO are the only alternative for solvent stabilization.

Residence Times

The coordination number discussed above is a static quantity. The corresponding dynamic properties concerning solute–solvent hydrogen bonds can be investigated using residence times in, for example, the first coordination shell. As described by Impey et al.,[21] we use the first hydration shell and then define a function $P_j(t,t_n; t^*)$ which takes the value 1 if the solvent molecule j lies within the coordination shell of a given solute atom at both time steps t_n and $t_n + t$ and in the interim does not leave the coordination shell for any continuous period longer than t^*. Otherwise, the function takes the value 0. As previously described, we set $t^* = 2$ ps[22] as well as $t^* = 0$ ps[23], i.e., in the latter case the first breakage is investigated.

We now define the function:

$$p(t) = \langle P_j(t, t_i; t^*)\rangle_{i,j} / \langle P_j(0, t_i; t^*)\rangle_{i,j} \quad (2)$$

which gives a probability for a solvent molecule in the coordination shell to remain there during time t. After a fast initial drop, the decay is almost exponential: $p(t) \sim \exp(-t/\tau)$, where τ is the residence time with τ_{decay} and τ_{simple} describing the two processes discussed. A summary of the calculated residence times is given in Table 3. In methanol all the τ_{decay} values are in the range of 10–20 ps. In contrast, there are large (up to an order of magnitude) differences in DMSO, especially for O6*m* with a short residence time and O2*g* with a long residence time. Analysis of τ_{simple} reveals that the values are on the short ps time scale and approximately one order of magnitude shorter than τ_{decay}, in both solvents.

Spatial Distribution Functions

To investigate the structure of molecular systems in liquids and solutions, radial distribution functions are clearly insufficient and sometimes even misleading. Particularly in cases where probable and nonprobable regions cancel each other in aver-

Table 2. Coordination numbers obtained from the radial distribution functions

Oxygen atom	n_{OO}-MeOH	n_{OH}-MeOH	n_{HO}-MeOH	n_{OO}-DMSO	n_{HO}-DMSO
O5*m*	0.4	0.0	–	0.6	–
O2*m*	1.4	1.6	0.7	1.2	1.5
O3*m*	1.5	1.5	0.9	1.1	1.5
O4*m*	1.5	2.0	0.8	0.5	0.8
O6*m*	1.0	1.8	0.2	0.3	0.4
O5*g*	0.4	0.2	–	0.4	–
O2*g*	0.8	0.6	0.6	1.0	1.9
O3*g*	0.4	0.0	–	1.0	–
O4*g*	1.4	1.9	0.9	1.1	2.2
O6*g*	1.3	1.7	0.9	1.0	2.0
O1*g*	0.5	0.3	–	0.8	–

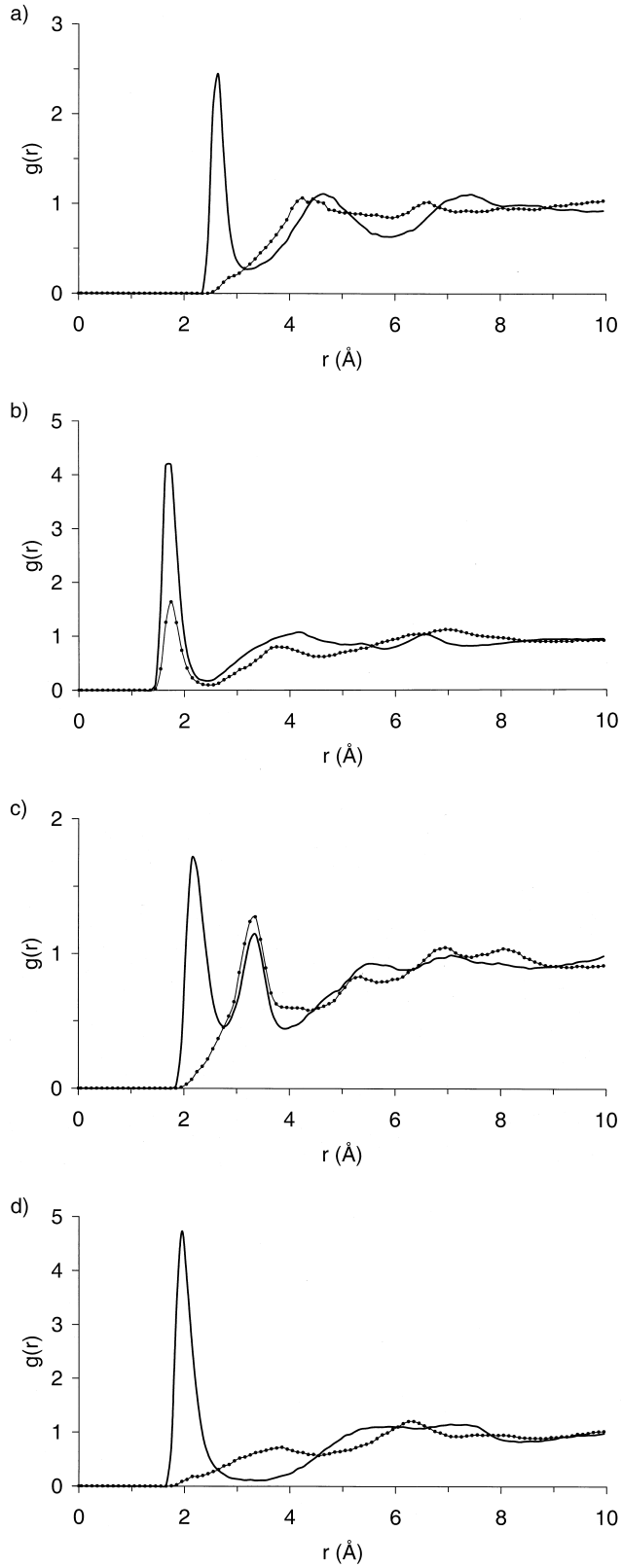

Figure 2. Radial distribution functions around the disaccharide: (a) O6m-O_{MeOH} (solid line) and O6m-O_{DMSO} (filled circles); (b) O2m-H_{MeOH} (solid line) and O2g-H_{MeOH} (filled circles); (c) HO6g-O_{MeOH} (solid line) and HO6m-O_{MeOH} (filled circles); (d) HO6g-O_{DMSO} (solid line) and HO6m-O_{DMSO} (filled circles).

Table 3. Residence times for hydrogen bonds between solvent and selected oxygens in the disaccharide. An oxygen-oxygen distance of 3.4 Å was used as a geometric threshold

Oxygen atom	Methanol		DMSO	
	τ_{decay} (ps)	τ_{simple} (ps)	τ_{decay} (ps)	τ_{simple} (ps)
O2*m*	12.0	1.8	11.7	1.4
O3*m*	10.9	1.6	15.9	2.6
O4*m*	12.3	2.0	7.5	1.0
O6*m*	13.3	2.4	5.5	0.5
O2*g*	10.0	1.9	42.9	5.3
O4*g*	18.7	3.1	31.5	4.9
O6*g*	15.3	2.3	34.2	5.3

aging over the angular coordinates. Detailed information about the three-dimensional solvation structure can be obtained using spatial distribution functions (SDF) of atomic density around a solute[24] given by:

$$g_{AB}(\vec{r}) = \frac{\rho_B(\vec{r}|\vec{r}_A = 0)}{\rho_B}, \qquad (3)$$

where similarly as in Equation 1, the first index A refers to a solute atom, the second index B refers to a solvent atom, r is a vector in cartesian coordinate space between the atoms, and ρ_B is the bulk solvent density of B in the solvent. SDFs give both the radial and angular dependence of the solvation structure around a solute (or part of a solute) fixed in the local coordinate system. The calculation of the SDFs can be done in a similar way as the RDFs during the simulations. In the case of large and flexible molecules, creating the local frame may become more difficult due to excessive intramolecular and conformational fluctuations as the simulation proceeds. Also, the visualization of SDFs creates some problems as these are four-dimensional quantities. The normal way is to freeze one of the dimensions by either using a projection onto two Cartesian dimensions or to use the isodensity surfaces of the SDF intensities, displayed in three dimensions. The latter method is used in this work and in our previous studies of the disaccharide α-D-Man*p*-(1→3)-β-D-Glc*p*-OMe in water and in water/DMSO solutions.[6] It should be stressed that SDFs should be used in combination with RDFs to have quantitative information, such as the coordination numbers, at hand.

In Color Plate 1, we see the SDFs in methanol solution. As expected, the solvation structure reflects the ability of methanol to both donate and accept hydrogen bonds. Besides, having in principle the same possibility to form hydrogen bond combinations as water, methanol can even hydrogen bond to the nonhydroxyl oxygens. The methyl groups in methanol impose restrictions in the solute–solvent hydrogen bonding pattern. The above analyses revealed hydrogen bonding that is readily identified in the SDFs, e.g. the donation of hydroxyl protons in methanol around O2*m* (upper left in Color Plate 1), O6*m* (middle right), and O4*g* (lower left) which was also shown to have the longest τ_{decay}. Clearly, the solvation structure of methanol around the disaccharide is not as extended as in water, but rather more like an average between that of water and that of DMSO presented below.

The most probable hydrogen-bonding sites for the DMSO oxygens around the disaccharide are depicted in Color Plate 2 using the same isodensity level. Distinct features showing hydrogen bonding to the hydroxyl groups are present. In a recent carbon-13 NMR relaxation study of disaccharides in DMSO, the rotational correlation time for the solute molecule scaled well with the number of hydroxyl groups in the disaccharide that could hydrogen bond to solvent.[25] Compared with our previous simulations of the disaccharide in water, where the corresponding SDFs showed distinct belt-like shapes around the entire solute, DMSO behaves differently. While four possible combinations of hydrogen bonds between the hydroxyl groups and water are conceivable, DMSO can only be engaged in one type of H bond. This is obvious if one analyzes for the three stabilizations in methanol discussed above, which are absent in DMSO. However, distinct regions are observed for HO4*g* (middle left in Color Plate 2), HO2*m* (upper right), and HO2*g* (lower right) also having the longest τ_{decay} in DMSO. Furthermore, the bigger size and much heavier mass slow down the mobility of DMSO and contribute to a more distinct type structure.

CONCLUSIONS

The MD simulations show that the conformation of the disaccharide is similar in the four different solvents studied, namely, water, water/DMSO 3:1, methanol, and neat DMSO. The combination of different types of analysis tools leads to a more complete picture of the solute in the different solvents, both with respect to static as well as dynamic structural parameters and reveals regions around the solute molecule where pronounced differences occur. Distinct regions of solvation are readily identified using spatial distribution functions as exemplified in the Color Plates in this article.

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25 Söderman, P., and Widmalm, G. Conformational flexibility of disaccharides investigated by multiple field

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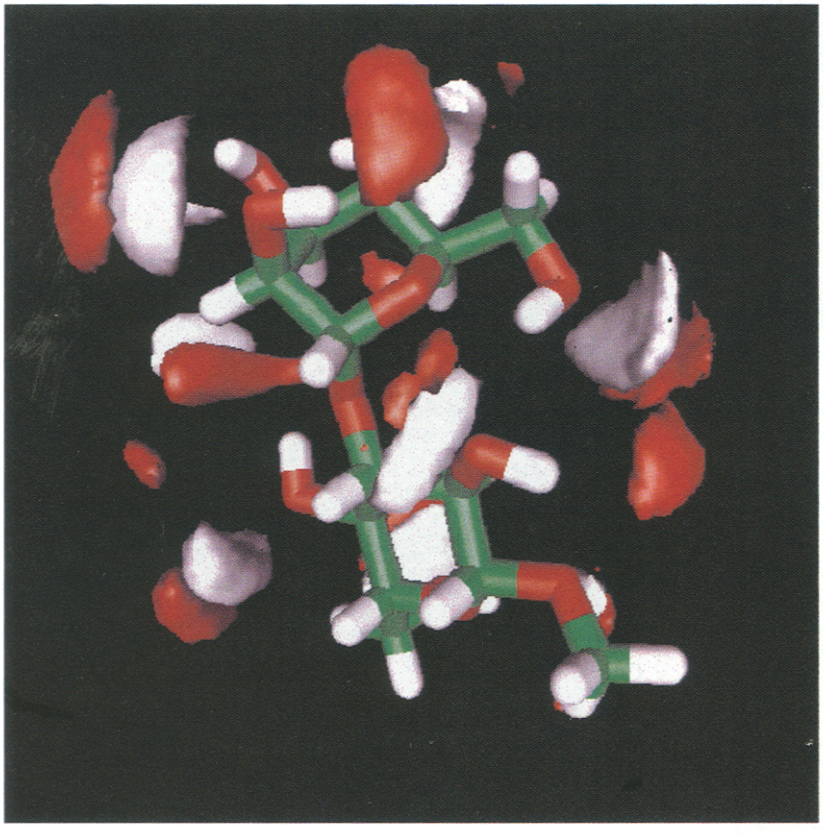
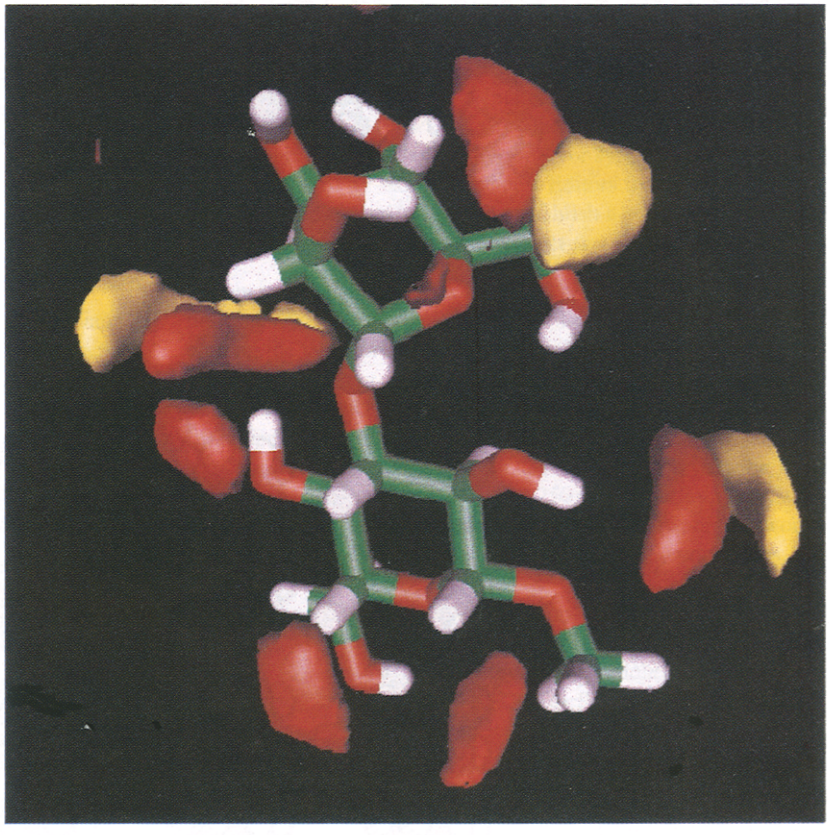


Color Plate 1. Spatial distribution functions around α-D-Man*p*-(1→3)-β-D-Glc*p*-OMe in methanol. Contours show the regions of high positive deviations of the local density (8.5 times higher than the average value). Atomic coloring is green for carbon, red for oxygen, white for hydrogen and yellow for sulphur.



Color Plate 2. SDF around the disaccharide in DMSO (the isodensity is 8.5).