Developing the Statistical Mechanics Theory of Liquids in Chemistry and Biophysics

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We have been exploring the chemical and biological processes in solutions, based on the statistical mechanics of liquids, especially, on the integral equation theory of molecular liquids or the "RISM" and "3D-RISM" theories.[1–3] Such exploration can be realized by combining the statistical mechanics theories with the other theoretical methods in the molecular science, which describes the different aspects of the physics such as the quantum processes and the liquid dynamics.

Our recent attention is focused on the "molecular recognition" and "fluctuation" of bio-molecules, which are the two key-processes in the living system. For examples, for an enzymatic reaction to take place, substrate molecules should be accommodated by the enzyme. The process is nothing but the molecular recognition which is regulated by the solvation free energy of the enzyme-substrate (ES) complex, and by the structural fluctuation of the protein.

1. Selective Ion-Binding by Protein Probed with the Statistical Mechanical Integral Equation Theory[4,5]

Molecular recognition is the most fundamental and important function of biomolecules. It is regarded as a process in which a host molecule makes a complex with a guest molecule through non-covalent chemical bonds including electrostatic, hydrophobic, and other interactions.

One of the most elementary processes of molecular recognition is the selective ion binding by protein. A variety of functions of protein is related to the ion binding: ion channels, ligand binding by a receptor, enzymatic reactions, and so on.

We have presented theoretical results for the ion binding by human lysozyme based on the 3D-RISM theory. The ion distribution around the wild type, Q86D, A92D and Q86D/A92D mutants in the several electrolyte solutions, KCl, NaCl and CaCl2, has been evaluated. The doubly substituted mutant, Q86D/A92D, has two isomers distinguished with whether it has a Ca^{2+} ion or not: apo, without Ca^{2+}; holo, with Ca^{2+}. Since the difference between wild type and mutants lies only in their active site, the discussion are focused on the active site, which consists of amino acid residues from Q83 to A92. The wild type and the Q86D mutant show no cation binding ability in accord with the experimental results. The 3D-RISM theory indicates that the A92D and Q86D/A92D mutants have cation binding ability. Na^{+} and Ca^{2+} ions are bound by the active site of the A92D and Q86D/A92D mutants, though K^{+} ions are not found in the active site. (see Figure 1)

The results are quite encouraging indicating the possibility of predicting protein functions by the theory. For example, it may become possible to find and/or design a protein, which has an ion binding ability. Such studies by means of the 3D-RISM theory are in progress.

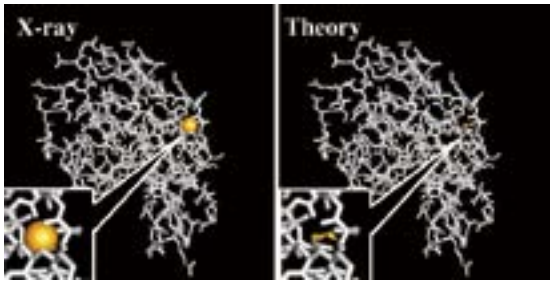

Figure 1. Comparison of Ca^{2+} position in holo-Q86D/A92D mutant between x-ray result and theoretical estimation. Threshold of 3D-DF is 25.0 for Ca^{2+} in the right hand side figure.

2. The Molecular Mechanism of the Pressure Denaturation of Protein Is Clarified by the 3D-RISM Theory[6]

It has been well regarded that protein denatures by applying pressure, but nothing is known to date about the molecular mechanism. The key to solve the question is the "partial molar volume (PMV)" of protein, because the volume should "shrink" in the denatured state of the molecule due to the Le Chatelier law. Therefore, the question "why and how does pressure change the structure of protein" can be rephrased as "why and how is the PMV of the high pressure structure (HPS)

less than that of the low pressure structure (LPS)."

In order to answer the question, we have calculated the PMV of a protein called "ubiquitin," associated with the transition from the low to high pressure structures, based on the 3D-RISM theory. The theory predicts that the PMV decreases upon the structural transition, which is consistent with the experimental observation. It is found from further analysis that the PMV reduction is ascribed substantially to the penetration of water molecules into a specific part of the protein. Based on the thermodynamic relation, this result implies that the water penetration causes the pressure-induced structural transition.

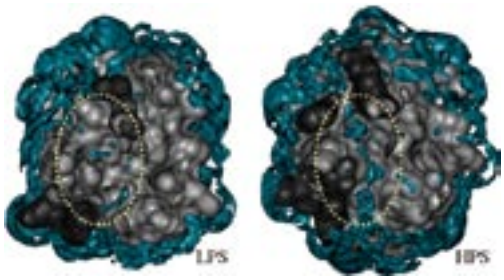

Figure 2. Isosurface representation of the three-dimensional distribution function of water oxygen around low-pressure (3 MPa) and high-pressure (300 MPa) structures (LPS and HPS, respectively) of ubiquitin. The blue surfaces show the area where the distribution function is larger than 2. The penetration of water molecules in the HPS is clearly seen in the area circled by the yellow dots.

3. Combination of Molecular Dynamics Method and 3D-RISM Theory for Conformational Sampling of Large Flexible Molecules in Solution[7)]

It has been a common understanding that the solvent plays an essential role in the thermodynamic stability of large molecules in solutions through, for instance, the hydrophobic and Coulomb interactions. In order to incorporate the solvent effects into the molecular simulations, we have developed a combination method of the molecular dynamics (MD) simulation with 3D-RISM theory. Using the proposed method, conformations of large flexible molecules in solution can be sampled along the free energy surface.

The solvent-induced force acting on solute atoms was evaluated as the gradient of the solvation free energy with respect to the solute-atom coordinates, which is obtained from 3D-RISM theory. In order to enhance the speed of computation, we have applied a multiple timestep algorithm based on the RESPA (Reversible System Propagator Algorithm) to the combined MD/3D-RISM method. To illustrate the present MD/3D-RISM simulation, we applied the method to a model of acetylacetone in aqueous solution, as a simple example of flexible solute.

To examine the validity of multiple timesteps, the dependence of energy conservation on timesteps was studied. Figure 3 shows the "time series" of the Hamiltonian H, where Δt_{RISM} denotes the timestep for performing the 3D-RISM calculation. The conformations of solute molecules were renewed with the timestep of 1 fs. For Δt_{RISM} = 1 and 5[fs], H is conserved within a tolerable accuracy. For Δt_{RISM} = 10 and 20[fs], however, the values of H deviate from their initial values, with "time" elapsing. The result indicates that we can choose the timestep Δt_{RISM} = 5[fs] without losing numerical accuracy: that is, the calculation of solvent-induced force once in 5 fs is sufficient for an accurate conformational sampling. This choice enhances the speed of computation by 3.4 times compared to a single timestep method, *i.e.* Δt_{RISM} = 1[fs].

Acetylacetone possesses an intramolecular hydrogen bonding capability between the hydroxyl group and the carbonyl oxygen atom, and the molecule is significantly stabilized due to this hydrogen bond, especially in gas phase. The intramolecular hydrogen bond was kept intact during almost entire course of the MD simulation in gas phase, while in the aqueous solutions the bond is disrupted in a significant number of conformations. This result qualitatively agrees with the behavior on a free energy barrier lying upon the process for rotating a torsional degree of freedom of the hydroxyl group, where it is significantly reduced in aqueous solution by a cancellation between the electrostatic interaction and the solvation free energy.

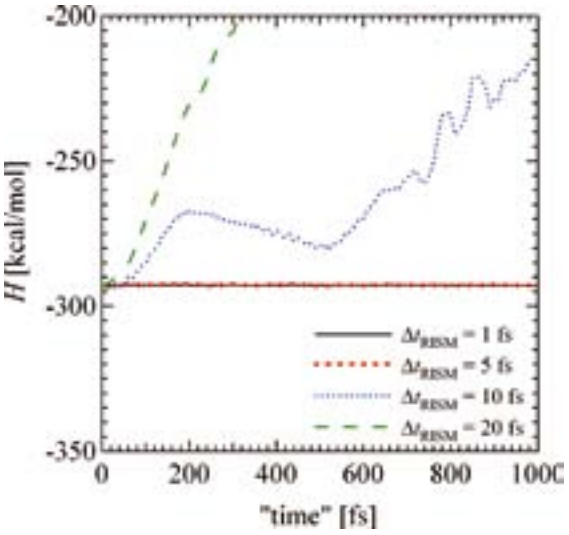

Figure 3. "Time series" of the Hamiltonian H. Δt_{RISM} denotes the timestep for performing the 3D-RISM calculation.

References

1) F. Hirata, *Molecular Theory of Solvation*, Kluwer; Dordrecht, Netherlands (2003).
2) A. Kovalenko and F. Hirata, *J. Chem. Phys*. **110**, 10095–10112 (1999).
3) T. Imai, R. Hiraoka, A. Kovalenko and F. Hirata, *J. Am. Chem. Soc. Communication* **127**, 15334–15335 (2005).
4) N. Yoshida, S. Phongphanphanee, Y. Maruyama, T. Imai and F. Hirata, *J. Am. Chem. Soc*. **128**, 12042–12043 (2006).
5) N. Yoshida, S. Phonphanphanee and F. Hirata, *J. Phys. Chem. B* **111**, 4588–4595 (2007).
6) T. Imai, S. Ohyama, A. Kovalenko and F. Hirata, *Protein Science* **16**, 1927–1933 (2007).
7) T. Miyata and F. Hirata, *J. Comput. Chem*. in press.