3E1034 粗視化 Go モデルを用いた Ferredoxin-like fold タンパクのフォールディング機構の解析**Analyses of folding processes on the Ferredoxin-like fold proteins by means of a coarse grained Go model****Masatake Sugita**, Takeshi Kikuchi (*Dept. Bioinf., Coll. Life Sci., Ritsumeikan Univ.*)

It has been recognized that analysis of proteins with a same native topology is efficient strategy for comprehending protein folding mechanisms. In particular, proteins sharing Ferredoxin-like fold are widely studied experimentally. It has been reported that these proteins exhibit various ϕ value patterns. These profiles are interpreted that stability of the two competing foldons regulates folding mechanisms. We are currently studying Ferredoxin-like fold proteins by analyzing amino acid sequence information. Our data also indicate that a significant segment for a folding is different among Ferredoxin-like fold proteins. For detailed elucidation of folding mechanisms with higher resolution including extensive free energy profiles, we have to conduct simulation approach. In this study, we analyze the folding processes of Ferredoxin-like fold proteins using coarse grained Go model. We use protein S6, U1A and ADA2h as target proteins. While we couldn't distinguish individual folding pathway using a topology-only model, we discern the difference of heights of free energy barrier. On the other hand, we got inherent folding process respect to each target protein using the model including sequence colors.

3E1046 異なる参照状態を用いた統計ポテンシャルによるタンパク質モデル構造の最適化**Optimization of protein model structures according to statistical potentials with different reference states****Matsuyuki Shirota**^{1,2}, Kengo Kinoshita^{1,2,3} (¹*GSIS, Tohoku Univ.*, ²*ToMMo, Tohoku Univ.*, ³*IDAC, Tohoku Univ.*)

Prediction of the 3D structure of a protein molecule from its amino acid sequence is an important challenge in computational biophysics. One of the bottlenecks is to select the most native-like structure among the model structures generated in the prediction process. Statistical potentials derived from protein structure database are widely used for this purpose. In principle, they are developed by counting the conformations in the database of native protein structures under the assumption that frequently observed conformations are energetically favorable ones. To quantify the significance of the counts of conformations in the database, the reference state, a virtual state in which all atomic interactions are random, is required. Previously, we demonstrated that the definition of the reference state of distance-dependent atomic pair statistical potentials can be theoretically variable and in practice statistical potentials with various reference states show different advantages, such as specificity for the native structures or the correlation to the native-likeness. To further explore the problem of reference states, here we developed an optimization method of protein model structures under several statistical potentials based on distinct reference states. In addition, we also employed several statistical potentials in a linear combination to improve the performance. Our results showed that various reference states have different advantages in both model evaluation and model optimization.

3E1058 クーロンレプリカ交換法を用いた A β フラグメントに関する研究**Studies on a A β fragment by the Coulomb replica-exchange method****Satoru Itoh**^{1,2}, Hisashi Okumura^{1,2} (¹*Institute for Molecular Science*, ²*The Graduate University for Advanced Studies*)

Effective samplings in the conformational space are necessary to predict the native structures of proteins. The replica-exchange method (REM) is one of the most well-known methods among the generalized-ensemble algorithms which realize effective samplings in the conformational space for biomolecular systems. However, REM has a difficulty for large systems that many replicas are needed to realize such effective samplings. In other words, huge amount of computer time is required in the replica-exchange method for the large systems. In order to overcome this difficulty, we proposed a new replica-exchange method, which we refer to as the Coulomb replica-exchange method (CREM), recently. In this CREM, electrostatic charges are exchanged among replicas although the temperatures are exchanged in conventional replica-exchange method. By exchanging the electrostatic charges, the free-energy barrier come

from the electrostatic interactions are reduced. Moreover, the number of replicas can be decreased by exchanging the electrostatic charges only of solute atoms. Consequently, the CREM simulations realize effective samplings in the conformational space with small number of replicas. I will show the effectiveness of this method through applications to a amyloid-beta fragment.

3E1110 レプリカ交換インターフェースプログラム(REIN)**Replica-exchange interface program (REIN)****Naoyuki Miyashita**^{1,2,3}, Suyong Re⁴, Yuji Sugita^{1,2,3,4} (¹*RIKEN QBiC*, ²*RIKEN CSRC*, ³*RIKEN AICS*, ⁴*RIKEN ASI*)

In the recent decade, replica-exchange molecular dynamics (REMD) method [1] has attracted much attention as an enhanced conformational sampling method. In REMD, ensembles consist of non-interacting copies (replicas) of an original system with different conditions. The pair of replicas are exchanged every few steps by metropolis criteria to enhance the sampling efficiency.

We have developed replica-exchange interface program (REIN) to facilitate REMD simulations. Currently, the multi-dimensional REMD can be performed [2] using the highly parallelized conventional MD (NAMD2 [3] and MARBLE [4]). The other conventional MD programs can be easily combined as well. REIN is also tested for use in supercomputer systems, enabling us to simulate substrate binding or conformational changes in large proteins with high precision. The detail of program and some recent applications will be presented.

[1] Y. Sugita and Y. Okamoto, *Chem. Phys. Lett.*, 314,141(1999)[2] Y. Sugita, A. Kitao, and Y. Okamoto, *J. Chem. Phys.*, 113, 6042 (2000)[3] J. C. Philip et al., *J. Comp. Chem.*, 26, 1781 (2005)[4] M. Ikeguchi, *J. Comp. Chem.*, 25, 529 (2004)**3F0900 サポート膜を用いた膜タンパク質の顕微鏡下でのイオン輸送活性計測****Evaluation of ion-pump activity of membrane proteins reconstituted on supported membrane under optical microscope****Jyunan Ki**, Rikiya Watanabe, Kazuhito Tabata, Hiroyuki Noji (*Department of Applied Chem., Univ. Tokyo*)

Supported membrane is the experimental platform in which a lipid bilayer is tethered on a solid surface. Owing to the mechanical stability, it allows us to investigate physicochemical properties of membranes and also membrane proteins reconstituted in the membrane. In this study, to enhance the versatility of this method, we developed a novel supported membrane system that enables to measure the ion-pump activity of membrane proteins under an optical microscope. As a model ion pump, we reconstituted F₀F₁-ATP synthase (F₀F₁) in liposomes. The supported membrane was prepared on a glass surface from the reconstituted liposome with a buffer containing a pH indicator dye (pHrodo) and caged-ATP. By introducing the evanescent illumination of 405 nm light for uncaging of ATP from the bottom of the glass surface, ATP was supplied only in the illumination area. The local pH of the membrane-surface space was monitored with the fluorescent signal of pHrodo excited with the evanescent illumination of 532 nm. Only after the illumination for uncaging, the intensity of pHrodo evidently increased, suggesting the acidification between the membrane-surface space. Furthermore, the degree of the acidification was proportional to the amount of uncaged ATP. These results showed that this systems allowed us to successfully monitor the H⁺-pumping activity of F₀F₁ reconstituted in the supported membrane.

3F0912 Catalytic activity of MsbA reconstituted in nanodisc particles is modulated by remote interactions with the bilayer**Takeaki Kawai**¹, Jose Caaveiro^{1,2}, Toyomasu Katagiri³, Hisashi Tadakuma¹, Takuya Ueda¹, Kouhei Tsumoto^{1,2,4} (¹*Grad. Sch. of Front. Sci., Univ. of Tokyo*, ²*Inst. Med. Sci., Univ. of Tokyo*, ³*Inst. Gen. Res., Univ. of Tokushima*, ⁴*Dep. Chem. Bio., Univ. of Tokyo*)

ATP-binding cassette (ABC) transporters couple hydrolysis of ATP with vectorial transport across the cell membrane. We have reconstituted ABC transporter MsbA in nanodiscs of various sizes and lipid compositions to test whether ATPase activity is modulated by the properties of the bilayer. ATP hydrolysis rates, Michaelis-Menten parameters, and dissociation constants of substrate analog ATP- γ -S demonstrated that physicochemical properties of the bilayer modulated binding and ATPase activity. This is remarkable when considering that the catalytic unit is located ~50 angstrom from the transmembrane region. Our results validated the use of nanodiscs as an effective tool to reconstitute MsbA in an active catalytic state, and highlighted the close