3P037 真菌由来エラスターゼインヒビター AFUEI と植物由来 potato I family インヒビターとの構造類似性
Structural similarity of AFUEI, an elastase inhibitor from *Aspergillus fumigatus*, and the potato I family inhibitors from plants

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AFUEI is an *Aspergillus fumigatus* elastase inhibitor that has a high inhibitory activity against elastases of *A. fumigatus*, *A. flavus* and human neutrophil elastase (HNE). Elastase from *Aspergillus* sp. is a major pathogenic factor for aspergillosis, therefore, AFUEI is one of the strong drug candidates of acute lung injury (ALI). We have determined the crystal structure of AFUEI and it shows remarkable similarity to serine protease inhibitors of the potato inhibitor I family, although their amino acid sequences are not similar. This similarity allowed us to build a complex model of AFUEI with HNE and suggests that AFUEI inhibits its cognate proteases through the same mechanism as the potato I family inhibitors.

3P038 酵母 26S および 20S プロテアソームの構造研究
Structural investigation of the yeast 26S and 20S proteasome

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The 26S Proteasome is a large multiprotein complex involved in a regulated degradation of ubiquitinated protein in the cell. It has not been the way itself how the 26S proteasome degrades ubiquitinated proteins. We have experimented that degradation process and/or particle forming by a small angle X-ray scattering. Scattering intensities were collected by use of a BioSAXS-1000 installed FR-E X-ray generator with a protein concentration of 1mg/mL. We have calculated a liberal structure of the 26S and 20S component without any constraints on shapes of components. We have purified another 20S particle whose fusion site was exchanged, and compared with a single cap 26S or a 20S core particle. In the session, we will discuss our strongly and weakly bound 26S characters.

3P039 *Mycoplasma mobile* の滑走時に“あし”として働くシアル酸レセプターの構造解析
Structural study of neuraminic acid receptor working as foot in *Mycoplasma mobile* gliding

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M. mobile glides on solid surface by a unique gliding mechanism. The "Leg" protein, Gli349 composed of 3183 amino acids has been suggested to have a neuraminic acid (Neu5Ac) receptor domain at its C-terminal part (foot). However, no similarity of amino acid sequence has been found to any other proteins. We predicted the foot structure based on the alignment of β -strands and found that the predicted structure is consistent with our current image from electron microscopy and has high similarity with toll-like receptor 4, involved in natural immune system. To elucidate the structure and functions of foot, we expressed and purified the foot composed of 465 amino acids using a synthesized gene and *E. coli* system.

3P040 藍色細菌時計タンパク質 KaiA-KaiC 相互作用の ESR 解析
Interactions between cyanobacterial clock proteins KaiA and KaiC revealed by ESR analysis

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The cyanobacterial circadian clock can be reconstituted *in vitro* from three clock proteins, KaiA, KaiB, and KaiC, in the presence of ATP. Here, we investigated interactions between KaiA and KaiC by the site directed spin labeling-electron spin resonance method, which can provide information on the location and environment of an individual residue within a protein. We labeled each cysteine residue of Cysteine-substituted mutants of KaiC and KaiA derived from the thermophilic cyanobacterium *Thermosynechococcus elongatus* with spin labels, performed ESR analyses, and identified specific residues involved in KaiA-KaiC interactions.

3P041 黄色ブドウ球菌の Isd-NEAT ドメイン間におけるヘム輸送についての考察
Insights into the mechanism of heme-transfer between Isd NEAT domains of *Staphylococcus aureus*

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IsdH-N3, IsdA-N, and IsdC-N are near transporter (NEAT) domains of iron-regulated surface determinant (Isd) proteins of *Staphylococcus aureus*. These domains bind heme and transfer it from IsdH-N3 to IsdC-N via IsdA-N in an affinity-driven manner. However, as the structures of the NEAT domains are quite similar to each other, it was difficult to explain the physicochemical basis of the difference in affinity.

To solve this problem, we performed MD simulations for these domains. They revealed important residues that have large contribution to the affinity. Then, we made mutant proteins with substitutions of these residues and measured their affinities for heme with ITC. We found that the mutations reduced the affinity for heme, as predicted by the simulations.

3P042 Hsp90 と ADP の解離過程における自由エネルギープロフィールと解離経路
Free energy profile and dissociation pathway in the dissociation process of ADP from Hsp90

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Hsp90 (Heat Shock Protein 90) is one of a group of molecular chaperones required for protein folding. The functional cycle of Hsp90 is driven by ATP binding, hydrolysis of ATP to ADP and ADP dissociation. The processes of ATP binding and ADP dissociation of are not revealed at atomic resolution. In this study, we performed all-atom molecular dynamics (MD) simulations of Hsp90 with ADP to calculate the free energy profile for ADP dissociation and to reveal the dissociation pathway of ADP. Thermodynamic integration method was applied to calculate the free energy profile for ADP dissociation. We will report the results of the free energy calculation and the possible pathway of ADP dissociation.