VII-H Development of Coordination Chemistry-based Strategies for Structural and Functional Modulation of Naturally Occurring Proteins or Enzymes

New methodologies based on coordination chemistry are actively developed which one can modulate or switch a structure and a function of naturally occurring proteins and enzymes. Using coordination chemistry on a specific site of a protein or peptide, we aim to explore a new functional molecules or chemistry-based strategy that can selectively recognize a specific surface of a protein and/or modulate its characteristic structure. Such molecules and methodologies would be expected to lead to functional change and regulation of complicated bio-macromolecules.

VII-H-1 Guest-Induced Umpolung on a Protein Surface: A Strategy for Regulation of Enzymatic Activity

HAMACHI, Itaru[1,2]; EBOSHI, Ryoji[1]; WATANABE, Jun-ichi; SHINKAI, Seiji[1]
([1]Kyushu Univ.; [2]IMS)

[*J. Am. Chem. Soc.* **122**, 4530 (2000)]

This manuscript describes a new strategy to introduce an on-off switch to an engineered enzyme. We successfully demonstrated that the transition metal ion-induced charge inversion (*i.e.*, guest-induced Umpolung) occurred on a surface of semisynthetic ribonuclease S' efficiently causes sharp on-off switching of enzymatic activity. Semisynthetic ribonuclease S' (RNase S'), an RNA hydrolyzing enzyme, was employed as a suitable model. As a surface charge modulator, an unnatural amino acid bearing iminodiacetic acid group (Ida^4) was incorporated into the S-peptide region of RNase S' by solid phase peptide synthesis. The mutant S-peptides were combined to S-protein by a self-assemble manner. The charge of the side chain of Ida^4 is a mono*anion* at neutral pH and it is inverted to a mono*cation* upon complexation with a trivalent metal cation such as Fe(III) at its iminodiacetic acid moiety. Based on the kinetic assay using single mutants, it is clear that the response to metal ions greatly depends on the charge of the original amino acid, that is, the Fe(III)-induced activity enhancement occurs at positively charged Lys or Arg site and the activity suppression occurs at negatively charged Glu or Asp site. A rationally designed double mutant displayed the sharp on-off switching (10–20 fold) of enzymatic activity which is sensitive to iron(III) concentration.

VII-H-2 Pd(en) as a Sequence-Selective Molecular Pinch for α-Helical Peptides

HAMACHI, Itaru[1,2]; KASAGI, Noriyuki[1]; KIYONAKA, Shigeki[1]; NAGASE, Tsuyoshi[1]; MITO-OKA, Yasuko[1]; SHINKAI, Seiji[1]
([1]Kyushu Univ.; [2]IMS)

[*Chem. Lett.* 16 (2001)]

A palladium(II) ethylenediamine complex was found to selectively stabilize a-helix conformation of peptides having two histidine (His) residues at *i* and *i* + 3 or 4 positions, whereas the helix conformation of the other peptides having one or two His at different positions is destabilized. Based on CD titration, NMR spectral observation and molecular modeling calculation, we established that Pd(en) is a versatile molecular unit for binding to peptides bearing two His at a specific pattern in aqueous solution. The ethylenediamine moiety in this organometallic receptor can be chemically modified with the combination to other binding interactions, so as to facilitate the more selective binding and modulation of a protein surface. This is sharply distinguished from the simple metal ions previously reported by other researchers.

VII-H-3 Zn(II) Dipicolylamine-Based Artificial Receptor as a New Entry for Surface Recognition of α-Helical Peptides in Aqueous Solution

MITO-OKA, Yasuko[1]; TSUKIJI, Shinya[1]; HIRAOKA, Takashi[1]; KASAGI, Noriyuki[1]; SHINKAI, Seiji[1]; HAMACHI, Itaru[1,2]
([1]Kyushu Univ.; [2]IMS)

[*Tetrahedron Lett.* **42**, 7059 (2001)]

Artificial receptors for bioactive peptides are actively developed in the field of the recent molecular recognition chemistry, because of their importance for the peptide sensing and pharmaceutical application. However, the recognition events have been exploited in organic solvents in the most cases, which are still far from the biological prospects. For the development of artificial receptors that can selectively bind a peptide/protein surface so as to inhibit or enhance the function, it is desirable to establish a design strategy for artificial receptors toward a peptide surface in aqueous solution. We describe herein that zinc(II) dipicolylamine (Zn(Dpa))-based coordination chemistry is promising for design of artificial receptors in aqueous solution toward α-helical peptides displaying two Histidine(His) on their surface (*H* – *i* and *i* + 4, or *H* – *i* and *i* + 7, or *H* – *i* and *i* + 11). The spatial juxtaposition by a modular connector greatly influences the affinity to these peptides. To the best of our knowledge, this is the first example for the artificial receptors that can selectively bind peptides bearing two His in the distance of two or three helix pitches in perfectly aqueous solution. This motif can be readily combined with other binding motifs, so that one can design the more selective and

efficient artificial receptors toward a peptide or protein.