論文の内容の要旨

Development of peptide-based covalent protein modifiers by means of *in vitro* selection

（標的タンパク質と共有結合を介して結合するペプチドの試験管内選択法による開発）

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Molecules that covalently bind to a native protein of interest are useful for detection and regulation of the protein. Although many such protein modifiers have been developed by embedding a reactive group into a non-covalent ligand scaffold, the conventional methodology often required intense efforts to achieve both selectivity and reactivity. In this thesis, I describe a new methodology for development of covalent protein modifiers with readily tunable reactivity by means of *in vitro* selection. In addition to a model target protein used for the establishment of the methodology, it was applied to another target protein to demonstrate the generality. In addition to the work on covalent modifiers, I also report mathematical modeling of receptor activation induced by dimeric macrocyclic peptides.

Chapter 1 is the general introduction to this thesis. It first describes selective covalent modifiers of native proteins focusing on conventional design strategies. Then, the chapter describes non-standard macrocyclic peptides and the *in vitro* selection system based on which the new methodology was developed. Finally, the purpose of the research in this thesis is briefly explained.

Chapter 2 describes the development of the new methodology for discovery of peptide-based covalent protein modifiers. I constructed libraries of macrocyclic peptides bearing reactive groups and developed an *in vitro* selection scheme for covalent modifiers and applied it to a model target protein. I also demonstrated that the reactivity of a discovered covalent modifier could be readily modulated by rational design.

Chapter 3 describes application of the newly developed selection scheme to epidermal growth factor receptor (EGFR). A selective covalent modifier was successfully identified, demonstrating the generality of the selection scheme.

Chapter 4 describes mathematical modeling of Met activation induced by dimeric macrocyclic peptides. To test the hypothesis that bell-shaped dose-response curves observed for receptor activation induced by dimeric macrocyclic peptide ligands could be explained by change in stoichiometry of the receptor-ligand complex, I constructed a mathematical model and compared it with the experimental data. The model could reproduce the observed profile and it was confirmed that the proposed stoichiometric change agreed with the model.

Chapter 5 is the general conclusion of this thesis. It summarizes achievements in this thesis and discusses perspectives.